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РЕЗЮМЕ

ФУЗАРИОЗНОЕ УВЯДАНИЕ У ПШЕНИЦЫ И ИНТЕГРИРОВАННЫЙ КОНТРОЛЬ БОЛЕЗНИ В РУМЫНИИ

И. МУНТЯНУ, Т. МУРЕШАН, В. ТАТАРУ

В статье сообщается о научных исследованиях и анализах 80 проб пшеницы, взятых из различных мест Трансильвании, а также о результатах некоторых моно- и полифакториальных опытов в отношении появления, поведения и снижения урожайности, вызываемой фузариозным увяданием. В 1970 году заболевание фузариозом было основной эпифитотией и причинило потери урожая в некоторых местах до 70 процентов, а в среднем для Трансильвании потеря урожая составила 42 процента. Потери изменялись в зависимости от сорта, большой изменчивости окружающей среды и условий агротехники. Высокие потери урожая наблюдались в случае обильного увлажнения, раннего посева в сентябре, избытка азотных удобрений, низкой устойчивости к полеганию, большого количества сорняков, раннего появления и сильного развития болезни. Это положение и наличие инфекции на растениях в течение вегетации, а также распространение ее с семенами должны учитываться специалистами при принятии специальных мер в отношении болезни, предпочтительно в качестве интегрированного контроля.

СПЕКТР ФЛУОРЕСЦЕНЦИИ АКТИНА

Ш. ФАЗЕКАШ, В. СЕКЕШИ-ГЕРМАНН, И. КАША, И. ХОРНЯК

В наших экспериментах изучалсъ спектры возбужденння и флуоресценции актина и изменение последнего. Спектры возбуждения и флюоресценции актина измерялись методом спектрофлюорометрии. Было найдено, что спектр возбуждения изменяется в большей, а спектр флуоресценции — в меньшей степени. Во время очистки актина ширина спектров с более гомогенной узкой полосой (290—310 мм) расширялась в спектре возбуждения в противоречие процессу очистки. Очистка, гельфильтрация, ультрацентрифугирование, полимеризация и деполимеризация, диализ приводят к гомогенному G-актину, что доказывается структурами ультрафиолетового спектра и спектром дифференция экстинция, увеличением полимеризационной способности, а также удаленными веществами. В противоположность этому расширение спектра возбуждения указывает не на стабилизацию, а на вторичное, третичное изменение структуры, на дезинтеграцию. Расширение спектра показывает, что имеется взаимосвязь между свободно и тесно связанными липидными компонентами, удаленными из протеина. Получены липидные компоненты гельфильтратного актина, их спектры возбуждения и флуоресценции, а также их тонкослойная хроматография. Липиды имеют смешенные компоненты.

АНАТОМИЧЕСКОЕ, УЛЬТРАСТРУКТУРНОЕ И ФИЗИОЛОГИЧЕСКОЕ ИЗУЧЕНИЕ ФОТОСИНТЕТИЧЕСКОЙ АКТИВНОСТИ, ОБНАРУЖЕННОЙ У КОРЫ EUONYMUS EUROPAEUS II; СЕЗОННЫЕ ИЗМЕНЕНИЯ

Ю. СУЙКО-ЛАЦА, Н. РАКОВАН, Г. ФЕКЕТЕ, Г. ХОРВАТ

Авторы изучали ультраструктурные и физиологические изменения хлоропластов в хлоренхиме одно- и двухлетних побегов **Euonymus europaeus L.** посредством электронномикроскопического исследования и ввения CO_2^{14}. Реакция пластов к тем же самым стимулам в данное время была не совсем одинакова, но обычно они поддерживали свою внутреннюю базисную структуру, требуемую для их активности в течение всего года. В пластинчатой структуре авторы обнаружили качественные и — что касается числа гранул — даже количественные сезонные изменения. Число гранул значительно уменьшалось в октябре и оставалось на низком уровне даже в январе; в марте и апреле — увеличивалось. Сезонное изменение общего содержания хлорофилла было параллельным с этим. Изучены эти две линии, поддерживающие одна другую. Годовое изменение фотосинтетической активности может быть параллельным с фенологией растений. В конце зимы фотосинтетическая активность очень высокая; в это время и в начале апреля синтез веществ с большим молекулярным весом может иметь значение в распускании почек. Во время активности листьев, активность коры уменьшается, биосинтез перемещается в направлении легкодоступных веществ с низким молекулярным весом.

ЯДЕРНАЯ ЗАВИСИМОСТЬ РЕСИНТЕЗА ГОРМОНОВ В АДРЕНОМЕДУЛЯРНЫХ КЛЕТКАХ КРЫС

И. БЕНЕДЕЦКИ, Л. КОППЕР, К. ЛАПИШ

Крысы обрабатывались 5-флюороурацилом или актиномицином Д для того, чтобы изучить функцию измененного аминокислотного обмена в процессе катехоламинного ресинтеза, следующим за инсулин-индуцированным катехоламиновым выделением. Инсулин (20 IE/100 г веса тела), введенный i. p. маркировал катехоламиновые выделения. Спустя 24 часа после обработки инсулином содержание катехоламина надпочечной железы равнялось только половине содержания в необработанном контроле. Через 168 часов после введения инсулина, уровень катехоламина возвратился к норме. Если 5-флюороурацил или актиномицин Д был введен через 6 часов после инсулина, ресинтез катехоламина был сильно угнетен. Так, через 168 часов адреномедулярное содержание катехоламина у повторно обработанных животных было равно только половине содержания инсулина обработанных контролей. Так как синтез гормонов имеет место в цитоплазме клетки и первичное действие 5-флюороурацила и актиномицина Д проявляется на клеточном ядре, то повидимому процесс синтеза цитоплазматических гормонов может нарушаться только в случае, если ядерный метаболизм является неповрежденным, т. е. процесс зависит от ядра.

ВХОЖДЕНИЕ ЯДРА ВЕГЕТАТИВНОЙ КЛЕТКИ, ГЕНЕРАТИВНОЙ КЛЕТКИ ИЛИ МИКРОГАМЕТ В ПЫЛЬЦЕВУЮ ТРУБКУ У CONSOLIDA AJACIS (L.) SCHUR.

ДЬ. ПАЛ, М. ТАЛЛЕР, Б. БАРНАБАШ

Когда начинается прорастание, ядро вегетативной клетки не всегда входит первым в пыльцевую трубку **Consolida ajacis (L.) Schur.** Генеративная клетка или две микрогаметы могут также входить первыми. Вхождение микрогамет в пыльцевую трубку и продвижение в ней при неактивном движении можно доказать. При дальнейшем росте пыльцевой трубки пыльцевое зерно становиться пустым и ядро вегетативной клетки, а также и микрогаметы переходят в каждом случае в пыльцевую трубку. В пыльцевом зерне и пыльцевой трубке микрогаметы изменяют место, по нашему мнению, посредством течения цитоплазмы вегетативной клетки микроспоры или посредством направленного и систематического сжатия цитоплазматических трубочек, включенных в цитоплазму.

ПОТРЕБНОСТЬ ДРЕНАЖА У РИСА

В. К. ВАМАДЕВАН, Н. Т. ДАТАНЕ

В оранжерее в экспериментах по дренажу наблюдалось, что и поверхностное осушение, и вертикальный дренаж оказали одинаковое влияние, но по их влиянию на урожай зерна и соломы оба они были более выгодными, чем обработка без осушения. При обоих обработках лучший рост растений можно отнести за счет большего количества продуктивных побегов и больших метелок.

ИЗУЧЕНИЕ МЕТАКСЕНИЙ У СОРТОВ ГРУШИ

Й. НЕКИ

В 1967—1970 гг. проведено изучение опыления у 645 комбинаций, каждый год с разным количеством сортов. В результате проведения помометрических измерений зрелых плодов, статистически доказаны метаксенийные изменения в форме и размере плодов, появлявшихся в течение трех лет, в следующих комбинациях: **Vilmos** (♀) × **Bosc** (♂), **Vilmos** (♀) × **Pringalle** (♂), **Vilmos** (♀) × **Dupuit** (♂); **Hardenpont** (♀) × **Pringalle** (♂), **Hardenpont** (♀) × **Vilmos** (♂), **Hardenpont** (♀) × **Clapp** (♂). Метаксении цвета плодов были получены в комбинациях: **Hardy** (♀) × **Vilmos** (♂) и **Hardenpont** (♀) × **Clapp** (♂). Что касается времени созревания, плоды в комбинации **Hardenpont** (♀) × **Pringalle** (♂) достигали уборочной спелости на 5—7 дней раньше, чем гибридные плоды всех других комбинаций.

ИЗМЕНЧИВОСТЬ СОДЕРЖАНИЯ ПРОТЕИНА И ЛИЗИНА В SORGHUM VULGARE.

А. АУСТИН, Г. Д. СИНГХ, В. К. ГАНСЛАС, Н. Дж. П. РАО

Девяносто шесть сортов сорго *(Sorghum vulgare)*, включающие местные и улучшенные сорта, а также гибриды, были оценены на содержание протеина и лизина в зерне. Показаны заметные сортовые различия по вышеуказанным признакам, так содержание протеина изменялось от 8,8%, до 21,0% и лизина от 0,72% до 3,37%. По содержанию протеина все сорта были разделены на низко-, средне- и высокопротеиновую группы. Шестьдесят три процента сортов относятся к высокопротеиновой группе. В сортах с низким содержанием протеина количество лизина изменялось от 1,24% до 3,37% при среднем, равном 2,21%, в то время как в высокопротеиновых сортах содержание лизина изменялось от 0,72% до 2,95% со средним, равным 1,52%. Высокодостоверный и отрицательный коэффициент корреляции между содержанием протеина и лизина в процентах (выраженный на 100 г протеина) показал, что в основном, увеличение протеина было связано с уменьшением содержания лизина в процентах. Однако имелись линии, такие как IS 4532 и IS 4952, которые содержали сравнительно больше лизина, сочетавшегося с более высоким содержанием протеина. Это определяет возможности улучшения линий, сочетающих высокое содержание протеина и лизина, обширно составленной программой и выбором подходящих методов селекции.

ИЗУЧЕНИЕ ФИЗИОЛОГИЧЕСКОЙ, А ТАКЖЕ БИОХИМИЧЕСКОЙ ОСНОВ КОМБИНАЦИОННОЙ СПОСОБНОСТИ ЛИНИЙ И ГИБРИДОВ КУКУРУЗЫ

М. КОВАЧ

Изучая 20 биохимических показателей в генеративных органах кукурузы, мы старались найти корреляцию между количеством отдельных составных частей и комбинационной способностью. В противоположность к предполагаемому положительному влиянию больших разниц между концентрациями мы наблюдали, что только определенная разница — около 10—20% — между родителями, использованными в скрещивании, вызывала получение успешных комбинаций. Однако, корреляция показывала только тенденцию, но не могла доказать прямого параллелизма. Положительная корреляция была выявлена между темпом размножения **Saccharomyces**, культивированных на семенном экстракте, и количеством урожая, также как между хорошей или плохой комбинационной способностью изученных линий.

ДЕЙСТВИЕ НЕКОТОРЫХ БИОЛОГИЧЕСКИ АКТИВНЫХ СОЕДИНЕНИЙ НА АЗОТНЫЙ ОБМЕН В МИЦЕЛИИ AGARICUS BISPORUS И COPRINUS COMATUS

Л. ДЬ. САБО, Л. ХОЛЛИ, Б. И. ПОЖАР

Авторы изучали действие нескольких групп биоактивных веществ (цитокинины, ауксины, гиббереллины, вещества, задерживающие рост и агенты старения) на развитие мицелия, содержание сухого вещества, общего и белкового азота у базидимицетов —

Agaricus bisporus — и **Coprinus comatus (Basidiomycetes).** Была обнаружена высокая биологическая активность у пуриновых и пиримидиновых аналогов, также как при влиянии обработками морфоактинами, Б-9 и этрелем; последние два вызывают старение у высших растений. В случае возбуждения также можно доказать эффективность, зависящую от концентрации. Селективность кофеина и фенобарбитуровой кислоты заслуживает особого внимания, так как вызывает увеличение сухого вещества у **Agaricus bisporus** и специфическое угнетение у **Coprinus comatus.**

МЕТОД КОНТРОЛЯ СТЕПЕНИ ПАСТЕРИЗАЦИИ В ПРОДУКТАХ ИЗ КОРОВЬЕГО И ОВЕЧЬЕГО МОЛОКА

А. ВАГНЕР

Автор рекомендует модифицированный бензидиновый тест, а также и быстрый фосфатазный тест, разработанный советскими экспертами и им самим, пироксидазный тест Сторха и Ротенфуссера (**Storch** и **Rothenfusser**) и тест Леверенца (**Lewerentz**) для контроля степени пастеризации молока и молочных продуктов и показывает, что совместным применением этих методов можно контролировать пятиминутную тепловую обработку при 65°C, а также и нагревание до 70°C, 75°C, 76°C, 78°C, 80°C и 90°C соответственно и наблюдать содержание сырого молока. Наряду с учетом результатов исследования подчеркивается важность производственного и технологического контролей. А с целью достижения безупречной эффективности пастеризации необходимо также избавить сырое молоко от маститис, как основного патологического материала, и от физиологически измененного, кислого и находящегося в стадии брожения молока.

ВАЖНОСТЬ УНИЧТОЖЕНИЯ САМЦОВ В ПРЕДОТВРАЩЕНИИ УЩЕРБА, ПРИЧИНЯЕМОГО КАЛИФОРНИЙСКОЙ ЩИТОВКОЙ QUADRASPIDIOTUS PERNICIOSUS COMST.)

Дж. ДЖЕНСЕР, Б. С. ИБРАГИМ

Под щитками и в стадии куколки самцы калифорнийской щитовки чувствительны к определенным ядохимикатам, как например к фосфористым органическим препаратам, испытанным также и в нашем институте. В течение развития перезимовавшей генерации—в противоположность условиям существования летнего поколения, — большинство особей внутри популяции находится на одинаковой стадии развития, самцы достигают состояния чувствительности к ядохимикатам в относительно короткий период времени. Подавляющее большинство самцов можно уничтожить в период начала взрослой стадии. В результате этого можно предотвратить массовое появление личинок, поскольку калифорнийская щитовка не размножается партеногенетически. Это мероприятие является важным прежде всего у тех косточковых, у которых появление личинок перезимовавшей генерации совпадает с созреванием плодов.

СОДЕРЖАНИЕ МИКРОЭЛЕМЕНТОВ В СЕНЕ ЛЮЦЕРНЫ

К. ПРОХАСКА

Изучалось содержание азота, а также основных макро- и микроэлементов в сене люцерны, полученного с покрытой песком почвы и чернозема. Данные показывают, что сено люцерны, выращенной на покрытой песком почве, содержало в каждом укосе меньше чистого белка и больше рядов волокна, чем выращенное на черноземе. Независимо от типа почвы, сено люцерны трехлетнего использования содержало меньше **Ca**, **K** и **Na**, чем сено двухлетней люцерны. Обнаруженные различия в содержании микроэлементов трехлетними растениями были недостоверны между двумя типами почв. Однако, содержание **Mn** и **Zn** в сене двухлетней люцерны было меньше. Отношение **Fe** : **Mn** : **Zn** : **Cu** :

Mo = 80 : 40 : 20 : 5 : 1, рассматриваемое как пригодное для корма не было достигнуто в исследуемом сене люцерны вследствие низкого содержания Mn и Zn, таким образом, принимая, что оно биологически неполноценное, можно оправдать дополнительное применение Mn и Zn.

ОПРЕДЕЛЕНИЕ ПЕРЕВАРИВАЕМОСТИ КОРМОВ ЦЫПЛЯТАМИ С ИСПОЛЬЗОВАНИЕМ ХИМИЧЕСКОГО МЕТОДА

АБД ЭЛЬ МЕГИД ДАРВИШ

Восемь опытов в трехкратной повторности по определению переваримости у петухов были предприняты для того, чтобы определить пищевую ценность корма, используемого в кормлении домашней птицы. Пять материалов были приготовлены раздельно, с тремя другими (мука семян хлопчатника, пшеничные отруби и высушенная люцерна) была использована кукуруза в качестве основного рациона. Для определения азота мочи, смешанной с экскрементами, был использован химический метод.

Здесь обсуждается коэффициент переваримости и кормовая ценность кормов, а также их отношение к исходным кормам.

РАСПРОСТРАНЕНИЕ НЕСИМБИОТИЧЕСКИХ АЗОТФИКСИРУЮЩИХ ОРГАНИЗМОВ В ПОЧВЕ В ОПЫТАХ С ДОЛГОСРОЧНЫМ УДОБРЕНИЕМ И СЕВООБОРОТОМ

А. Н. ИБРАХИМ

Несимбиотические азотфиксирующие организмы, названные *Azotobacter* и *Clostridia* были подсчитаны в почвенных образцах, собранных с различных экспериментальных участков с постоянным удобрением, расположенных в Бахтэеме. Азотобактер был обнаружен в более высокой плотности, чем *Clostridia.* Органическое удобрение достоверно увеличивало популяцию как азотобактера, так и *Clostridia* и производительность азотной фиксации была в этом случае намного выше, чем при внесении полного минерального удобрения. Система севооборота, кажется, не оказывала влияния на такую активность, но тем не менее участки с трёхгодичным севооборотом показали отчасти более высокую активность.

ВЛИЯНИЕ ДОЗЫ ГАММА-ОБЛУЧЕНИЯ НА РОСТ И МЕТАБОЛИЗМ ФОСФОРА И СЕРЫ У ГОРОХА

Й. ФРАНК, Л. ТОЛНАИ

В растениях гороха, облученных дозами 0,5, 1,0 5,0, 10,0 и 20,0 кр. на третьей неделе после прорастания увеличивается поглощение неорганического PO_4^{32} и его проникновение в органические фракции. Органические соединения малого молекулярного веса при дозе облучения 5,0 кр. составляют исключение. Стимуляция имеет свой максимум и при малых и больших дозах. Подобная тенденция проявляется и при применении SO_4^{35}, с той разницей, что меньший максимум большей дозы появляется при 10,0 кр., а при 20,0 кр. уже можно наблюдать его уменьшение. Гамма-облучения (за исключением стимуляции три 0,5 кр.) имеют задерживающий эффект во время морфогенеза растений, при этой депрессии выявляется максимум влияния меньшей (1,0 кр.) и большей (20,0 кр.) дозы облучения на длину стебля.

СПЕЦИФИЧНОСТЬ СТИМУЛЯЦИИ РОСТА, ВЫЗВАННАЯ ВОДНЫМ ЭКСТРАКТОМ ГАЛЛОВ У *Salvadora persica L.*

К. Д. ШАРМА, Д. Н. СЕН

Salvadora persica — растение пустыни, которое образует галлы почти на всех своих частях. Эти галлы, как здесь было показано, имеют не только вещества, задерживающие, но и стимулирующие рост. Водяной экстракт галлов оказал специфически различное стимулирующее влияние на рост двух выбранных для настоящего исследования видов *(Pennisetum typhoideum* и *Asterantha longifolia)*. Повидимому имеется больше чем одно стимулирующее рост вещество — как показано их различным значением Rf, — но только несколько из них были достойны упоминания. Эти стимулирующие рост вещества оказались теплостабильными, т. к. стерилизация экстракта не повлияла на их качество, в то время как задерживающие рост вещества были термолабильными.

ИЗУЧЕНИЕ ОТНОШЕНИЯ МЕЖДУ НЕСПОСОБНОСТЬЮ К ПРОРАСТАНИЮ И АКТИВНОСТЬЮ ЭНЗИМА ДЕГИДРОГЕНАЗЫ В СЕМЕНАХ ГОРОХА

К. ЛАСЛО

Изучая причину неспособности к прорастанию с точки зрения удовлетворительной или неудовлетворительной активности энзимов, катализирующих физиологические процессы прорастания, автор обнаружил корреляцию между экстрагируемым, экстрагируемыми + связанными и топохимически определенными активностями дегидрогеназы в семенах с одной стороны, и способностью к прорастанию, в %, выражающей отношение между прорастающими и непрорастающими семенами, с другой стороны.

РАЗЛИЧИЯ МЕЖДУ TEXAS (TCMS) И USDA (SCMS) ТИПАМИ ЦИТОПЛАЗМАТИЧЕСКОЙ МУЖСКОЙ СТЕРИЛЬНОСТИ

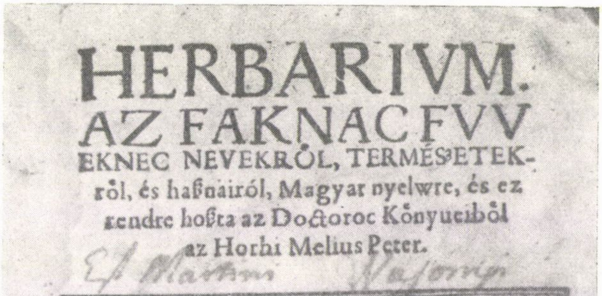
Ф. Т. ОРАБИ

Значительные различия наблюдались между цитоплазматической мужской стерильностью типов **Texas** и **USDA** как в Мартонвашаре, так и Сентлёринце по высоте початка и по числу листьев над верхним початком, и только в Сентлёринце по урожаю зерна. Не было значительного различия между двумя типами по высоте растений, ни в Мартонвашаре, ни в Сентлёринце. Эти два типа цитоплазмы взаимодействуют с данным генотипом разным способом. Техасская мужская стерильность отличается в значительной мере от мужской стерильности **USDA**. Условия среды оказывают значительное влияние на восстановление фертильности.

СУТОЧНЫЙ И ГОДОВОЙ РИТМ ВОДНОГО РЕЖИМА У СОРТОВ ПЕРСИКА, СОЗРЕВАЮЩИХ В РАЗНОЕ ВРЕМЯ

ДЬ. БОРКА, К. БОРКА

Относительно водного режима сортов персика были сделаны следующие наблюдения. Транспирация более интенсивна у более молодых листьев. В начале вегетационного периода, при одинаковом запасе воды, транспирация наиболее интенсивна у сорта Юлия. В дневном ритме транспирации сорт Юлия первым достигает максимума, к полудню транспирация уменьшается у всех трех сортов вследствие отсутствия тургора. Дефицит насыщенности водой, измеряемый в разное время дня, имеет обратную зависимость с водным потенциалом. В полдень, при максимальной инсоляции, температура листьев, страдающих от дефицита насыщения водой, на 4—5° С выше, чем на стадии полного тургора. Вследствие временного дефицита воды, происходящего в листьях в результате увеличенной транспирации, количество свободной воды уменьшается и увеличивается количество связанной воды. Имеется отрицательная зависимость между густотой и размером устьиц на листьях у изученных сортов. Чем ближе содержание воды в почве к ее общей водоемкости, тем выше число открытых устьиц.



HERBARIVM.
AZ FAKNAC FVV
EKNEC NEVEKRŐL, TERMÉSZETEK-
ről, és haſznairól, Magyar nyelwre, és ez
rendre hoſta az Doctoroc Kőnyueiből
az Horhi Melius Peter.

Nyomtattot Coloſuárat Heltai Gaſpárne
Műhellyébé, 1. 5. 78. Eſztendőben.

PIIS · MANIBVS

PETRI • MELII • JUHÁSZ

MDXXXVI—MDLXXII

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REDACTORES

ACTORVM · AGRONOMICORVM

ACADEMIAE · SCIENTIARVM · HVNGARICAE

WE DEDICATE THIS COPY AS A TOKEN OF REMEMBRANCE TO
PÉTER MÉLIUSZ JUHÁSZ (1536–1572)

The Editors

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PÉTER MELIUS JUHÁSZ

(1536?–1572)

The birthplace of Hungarian botany and the research of the wonderfully rich Hungarian flora is the town of Debrecen. The first definite data on the country's flora are the local names of Debrecen. Such names are found in the first Hungarian botanical — and at the same time medical and agricultural — work, the posthumous book of Péter Horhi Melius Juhász, the great reformer, one of the outstanding figures of the 16th century. The title of the book is: Herbarium. Az Fáknac Füveknec nevekről, természetekről és hasznairól. Magyar nyelwre, és ez rendre hoszta az Doctoroc Könyueiből az Horhi Melius Peter. (Nyomtattot Colosuárát Heltai Gáspárne Mühellyebé, 1.5.7.8. Esztendőben.) Little 8° 188 pag. (Herbarium. On the names, characters and uses of trees and grasses. Selected and translated from medical books by Peter Horhi Melius. (Printed at Kolozsvár, in the printing house of Mrs Gáspár Heltai, 1578.)*

The ardent preacher and bishop of Debrecen, who with his invincible belief and conviction, extensive knowledge, excellent organizing ability and strong will made Debrecen the Calvinist Rome and was called Pope Peter by his opponents, found time and felt enthusiasm even amidst his fights and struggles to give medical instructions to the poor Hungarian people. Namely, Melius' Herbarium is in fact a medical guide which explains how to use the plants listed for popular drugs.

At that time the naturalism of the Renaissance created a new era in botany. The fantastic and mythical specimens of primitive plant culture together with the beliefs and legends attached to them were pushed into the background by new books (the Herbarius or Kräuterbuch) on medical botany published mostly in Latin or German, which gave more and more perfect descriptions and naturalistic and artistic pictures — the drawings of plants, especially in the form of xylography reached a zenith in Dürer's century —

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* Indices before the text (Latin, Hungarian and German plant names as well as the list of diseases) and his letters are written in Latin capital letters.

and discussed the curing ability, useful or harmful nature of plants in detail. These books served as sources four Melius who — though from a scientific historical point of view his book can only be considered as a Hungarian transplantation — was the first Hungarian representative of botany and medical science.

P. Melius Juhász was born in the thirties of the 16th century, probably in 1536 at Horhi, a village of County Somogy destroyed later. (Thus, his numerous works must have been produced, his important social and political actions carried out in 36 years, which is hardly believable.) Of his origin and youth nothing is known. His name Melius — assumed according to the humanist practice — is the latinized form of the Greek Melios which means shepherd. In 1553—54 he went to school at Tolna; in the autumn of 1556 was student at the University of Wittenberg — it was there that he got acquainted with the herbals and began to translate them —; in 1558 was invited by János Enyingi-Török to be a priest in Debrecen, then the town itself elected him pastor and later bishop. He became the leader of the feudal-bourgeois, clerical and social life of the town, it was under his influence that people in the Great Hungarian Plain and partly in Transylvania became Calvinist. All his life he was an enemy of the Catholic Church, but was hostile towards some protestant sects too, like Lutheranism and especially Unitarianism. He attacked the Unitarian leader Ferenc Dávid fiercely (once in 1569 at Nagyvárad, in the presence of János Zsigmond Prince of Transylvania, who reproached him for it too). He strongly believed he was right; he was rigorous and dogmatic. Within eleven years he published 28 works. Latin and Hungarian polemical and theological writings, sermons difficult to read today. His translation of the Bible have got lost, so the most durable reminder of his activity is the Herbarium.

It is not known whether the manuscript of the complete work was found among his papers after his death or — as supposed by Natter-Nád — it was the editor who compiled it from his writings. The book was possibly written in four parts, and while at the beginning it is almost a word for word translation of Lonicerus' "*Naturalis historiae opus novum*" (I—II. Frankfurt, 1551, 1555) and "Kräuterbuch" (Frankfurt, from 1557 on in numerous editions, Gombocz uses the 1569 edition for comparison), later other authors too are often cited. In fact the Herbarium remained unfinished; it consists of 232 chapters, mentions about 620 plant species with some two thousand names of plants, but the "Kräuterbuch" contained 419 chapters. Of the contemporary herbals Melius certainly used the works of Fuchs, Matthiolus and Bock (Tragus), sometimes even criticized them (pag. 59a, 80a, 182y, 187a) and protested against the delusions and superstitions of the ancients. He said about one of Plinius' statements: "it is false idolization against God". Melius himself was a practical botanist, mentioned the source of origin of a number of plants in the Nyírség (region in the north-eastern part of Hungary) ("Nyíri föld") and in the neighbourhood of Debrecen (the names of "Csere, Úrréte, Fancsika, Bodóháza, Malomgát, Pércs" etc.); these were the first concrete data on Hungarian flora. His medicinal indications were partly tested by himself ("Próbált dolog") (Things tested). He adopted a number of plant species unknown in Hungary from Lonicerus or other authors; although he mixed up many similar or related species and genera, such mistakes can be found in other contemporary herbals too. Each chapter contains the Latin, Hungarian and German names of the discussed species, and sometimes some of their characteristic features; if more than one plants are discussed their distinctions, and seldom their habitat. Under the title "Natural" the wetting, drying, heating or cooling nature of plants, in the chapter "Inward uses" their internal, while in that of "Outward Uses" their external uses are discussed. At the end of certain chapters even formules of oils, syrups to be prepared from the plants in question are given. At the beginning of his work Melius discusses — relatively briefly — the woody plants; fungi and mosses are mentioned only in one chapter each (De Fungis, De Musco); the greater part of the book contains the "names and characters of grasses" meaning by it the herbaceous flowering plants; in the chapter "De Gramine" the sedge family is dealt with very briefly. Beside the numerous — sometimes a bit magical — formulas the book gives much practical advice too; so it gives instruction in the treatment of wine, preservation of meat, control of swine-fever, in killing bugs, lice, flies, mice, rats, bats; recommends medicaments against bee-sting or baldness; it even teaches ink preparation and book painting. It is remarkable that it recommends very different plants for the same illness; these are — of course — mostly ineffective. Melius sometimes adopts the natural history fables of the classical authors. Nevertheless, a very great part of the identified plants was used even in the last century, at least in popular medication; some 140 of them are well known Hungarian medicinal plants even today, and 62 are included in the official dispensatory. According to Halmai 72.6 per cent of the Hungarian medicinal plants can be found in the Herbarium even if their role and importance was then not the same as today.

Lajos Fialowski's study of Melius (1885) had been lost, so Endre Gombocz (1936) identified Melius' plant names with the present nomenclature; with 394 species he achieved a fairly reliable result. Natter-Nád completed them with names (1962) that are either totally unreliable or wrong (often they are not even correctly written), the author was apparently no expert. The greatest merit of the Herbarium is its recording nearly two thousand old Hungarian plant names; it is in this respect the most important source beside Balázs Szikszai-Fabricius' Nomenclature written in the middle of the century and published in 1590. It is a pity that many of its good Hungarian names were replaced by the awful words of the language reform. The value and usefulness of the book is indicated by the fact that more than one epigon and plagiarist works were published. E.g. András Beythe's "Fives könyv" (Book of herbs) (1595, Németújvár) is partly a translation of Matthiolus (*Commentarii . . .*) but mostly a copy of Melius. Melius presents valuable data on the old Hungarian names of diseases too; e.g. "francu": syphilis, rászt, now: lépdaganat — tumour of spleen, rothasztó feneseb, now: rák = cancer, tágy, now: kelés = boil, mérges köves kelés, now: furunkulus = furuncle, etc.; he sometimes describes even the symptoms of the diseases.

Melius' book is mentioned in Pritzel's large bibliography (ed. 2. 1872: 159) though with the title cited incorrectly and the author's name written Juhász vel Ihász. Here it mentions the name of Horányi (Ferenc Elek) as mentioning a 1662 edition of the Herbarium in Debrecen in his work on literary history (1775—77); such an edition never existed. Pritzel cites, further, the only Bohemian herbal published in the 16th century; it is a translation of Matthiolus' classical work *(Commentarii ad Dioscoridem . . .)*, a beautiful artistic work with fine xylographs, in two editions, both published in Prague under the title Herbarz: ginak Bylinarz . . ., in 1562 (by Thaddeus Hagek) and 1566 (by Adam Huber and Daniel Adam). The former — which is in the possession of the author of this paper — contains the Czech, Latin and German names, pictures, short descriptions and medical relations of plants. Other herbals of those times are not known in the socialist countries.*

Melius' Herbarium was published by Gáspár Heltai's widow. May this commemoration on the 400th anniversary of Melius' death be concluded with a few lines of the Preface. "These were the things the wise man Peter Melius, pastor of the Christian Church of Debrecen racked his brain about in our days. He was busy at collecting data on medicines for common diseases and translating them into Hungarian. His was the trouble of collecting them from books of various wise doctors and to describe them, mine was the work and expenses of publishing. May the Hungarian nation accept it kindly from me, poor widow."

R. Soó

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* The author of the present paper possesses an incomplete copy of Melius' book, various editions of herbals from Fuchs, Matthiolus, Bock, moreover, as incunables the Hortus sanitatis minor, 1493 (with mere descriptions and xylographs of plants) and Hortus sanitatis major, 1497, which contains animals and minerals too.

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THE HERBAL OF PÉTER MELIUS JUHÁSZ AND HUNGARIAN MEDICINAL PLANTS

The character of Péter Melius Juhász, his life and activity, the birth of the "Herbarium" and some of its popular therapeutic implications, the plants contained in it, the value and effect of the book have been discussed, criticized and appreciated by many authors. Little has been said, however, about the medicinal plants included in the book.

A work and its author can be properly and objectively criticized by a later age only in connection with their own age and circumstances. This method was chosen by the author of the present paper too when evaluating the medicinal plants from a medical-botanic i.e. therapeutical point of view. It is, of course, of considerable interest, which of Melius medicinal plants are known today, what they are used for, and what they were used for by the medical science of his time. Since Melius wrote his book for common people, the question arises what the people, and what the official therapy of that time as well as of later ages adopted from it. We ought to know whether from a modern viewpoint Melius' teaching was mere superstition and quackery, or represented the medical science of that time, and finally, how modern medicine evaluates the medicinal plants contained in Melius' book. And — when all this has been considered — whether the "*Materia medica*" and "*Ordinatio*" were progressive and of a scientific level?

These are important questions to which the answer can only be given after detailed analyses, thorough professional studies, taking the distance in time in consideration and with objective criticism. Time and space are not sufficient here to solve these questions. Nevertheless, this paper presents a number of data and will draw some conclusions at the end. A plant *(Agrimonia)* is used as a model to present the objective of the author's earlier work.

At that time, and even later, botany was practised for the purpose of curing, mainly by doctors and some natural scientists, but also by priests and preachers who had the duty of taking care of and curing both soul and body. This was Melius' aim too in publishing his Herbarium.

In an earlier work (HALMAI 1962) the author of the present paper gave — on the basis of Gombocz's identifications (GOMBOCZ 1936) — the Latin names of the plants contained in the Herbarium, their synonyms, and described the diseases they were used for, on the basis of literary data collected up to the end of the 19th century (DRAGENDORFF 1898). Plants employed in popular therapy in the middle of the century as well as those occasionally used abroad are also named (AUGUSTIN—JÁVORKA—GIOVANNI—ROM 1948); it is pointed out, further, whether they are included in the Hungarian dispensatories and among the latest Standard Prescriptions of that time (Fo-No), and the descriptions are completed with the domestic occurrence of the plants (JÁVORKA 1925); finally, their present use in popular therapy is also touched upon (OLÁH 1961).

The diseases, their curing and mainly the medicines of plant origin contained in the "*Pax Corporis*", a work written by Ferenc Pápai Páriz, teacher of the famous Nagyenyed College and physician of the Prince of Transylvania provided an interesting material for comparison with Melius' book.

Pápai Páriz' work was published 110 years later than Melius' book; the two served the same purpose: to restore the health of the people; the latter, however, was written by no clerical man but by a highly esteemed physician. In the author's above mentioned work 355 medicines of plant origin included in the "*Pax Corporis*" are described too.

Melius described 385 flowering and 8 thalloid plants in his work, and mentioned a moss too, by the name muscus sp.

As an example the text written on *Agrimonia* will be presented here: *Agrimonia eupatoria* L. herb, for healing wounds, internally, against liver pains, diarrhoea, intestinal parasites, and as a gargle against pharyngitis. (Drag. 280.) In Fo-No IV. its herb is a component of *Species Agrimoniae composita (cholagogum)* and *Species frangulae composita (laxativum)*. It is used internally in intestinal, lung and liver troubles, against gall stones; externally in hepatitis, thrush, and for rinsing mouth and throat (Aug. 132). — in Oláh's collection.

In Melius' work there are more than 600 plant species and some 2000 plant names. 394 of them were enumerated and 384 species identified by Gombocz; 7 were dubious and with 4 of them only the genus was clear (GOMBOCZ 1936). Some of the plants were of a foreign origin (*Aloe*, *Capparis*, *Cassia*, *Cedrus*, *Chrysanthemum*, *Citrus*, *Guajacum*, *Mandragora*, *Nardostachys*, *Olea*, *Phoenix*, *Punica*, *Rheum* etc.). This rich material proves that the author was familiar with a lot of contemporary works related mainly with the medicinal plants of Central Europe, and was familiar with the Hungarian flora as well.

The main lines and method of preparation of the author's earlier work will be presented here in some detail in order to support the conclusions drawn at the end.

In the list after the Latin names of the plants data on their use are given on the basis of DRAGENDORFF's (1898) work. In this highly valuable book the therapeutical application of some 13000 plants is discussed on the basis of data published till 1898. The author compiled written relics from the earliest times with thorough care, and gave data, sometimes even criticism concerning the active agents and the historical relations. This work was thus exceptionally suitable for comparing with Melius' material so that the latter could be evaluated after four hundred years.

Dragendorff generally gives the diseases by their Latin names. E.g.: *amenorrhoea* (absence of menstruation), *aphonia* (loss of voice), *arthritis* (gout), *catarrhus* (catarrh), *flatulentis* (suffer from flatulence), *dysuria* (retention of urine), etc.

It was difficult to decide how to give the names of the diseases: Latin names sound more scientific but are understood by a limited number of people, and the comparison with Melius' text becomes difficult, if designated by their Hungarian names they are understood by many, comparison is easy, and the whole book is given a popular character. For this, and for some other reasons too, the latter was the solution chosen.

1) The Herbarium was made for the people. Melius wrote it in the people's language, therefore we too have to comment upon it, complete and compare it in the people's language. 2) Certain diseases are no longer referred to by their Latin names, e.g. *Phthisis* (consumption, probably TBC), *Lithiasis* (stone troubles), *Konfortativum* (roborant or stimulant, very seldom used), or *Resolvens resolvent*, *Emmenagogum* (a drug promoting the bleeding of the uterus), while their Hungarian names are well known by the people even today. 3) Some of the Latin names were kept, as they are generally known e.g. antiseptic, diphteria, gonorrhea, syphilis, tetanus. 4) It was duly emphasized that on the basis of what is described nobody should cure him/herself or other people; only qualified persons should do that. Namely, catarrh, fever, bloody discharge or bloody sputum are not only initial signs of a simple disease, but often symptoms indicating the final stage of some serious or even mortal illness (TBC, cancer, etc.). 5) Finally, it was also pointed out what a large number of diseases we are endangered by, and incompetent people are liable to generalize. For example, catarrhs are of many kind; stone troubles may originate from the diseases of the bile, kidney, bladder, ureter, not to mention the innumerable illnesses of the uterus, the chest as well as the influence of character, physique and time.

It was also made known whether any part of the medicinal plants were included in the official Hungarian pharmacopoeia (fifth edition published in 1955); and references were made to the Standard Prescriptions (*Formulae Normales* — Fo-No) published on 1958. This latter, namely, was progressive enough to make 15 tea mixtures containing 18 new medicinal plants official. The fifth edition of the Pharmacopoeia contains 96 medicinal plant paragraphs. The data of these two sources reflect the official acknowledgement of the medicinal plants by physicians. They are completed with data from AUGUSTIN—

Jávorka—Giovannini—Rom (1948); in their work 196 Hungarian medicinal plants are discussed in detail and 42 briefly, completed with their popular and foreign utilization.

These three sources hallmark the medicinal plants of Hungarian origin, the number of which has even grown since then. Medicinal plants applied in popular therapy may be 1000 in number (Halmai: Magyarország gyógynövényei [Medicinal plants in Hungary]. Gyógyszerészek Lapja, 1936. 19. in series). With the data cited, collected by Andor Oláh dr. in county Békés a deeper insight into popular therapy can be obtained.

Last but not least Jávorka's work has to be mentioned (1925). There were two reasons for citing from his data. One of them was to prove the mainly Hungarian origin of Melius' material. The second was the fact that this work dealt with Hungarian plants in Melius' days, while recent works on the Hungarian flora with those in our days, and the two are enormously different. Thus, if the book published in the 16th century is considered, it can be done only in relation to the contemporary conditions, otherwise quite false results are obtained.

As a final conclusion about 480 of the 600 medicinal plant species included in Melius' Herbarium were found to be successfully identified (Gombocz). There are only 4 *Ranunculus* species the therapeutical utilization of which has not been discovered in the literary sources mentioned, all the others — 99 per cent — are registered as medicinal plants. 62 of them are included in the Hungarian dispensatories and the *Formulae Normales*, that is, are legally accepted by official medicine after some 400 years, and 138 are acknowledged as Hungarian medicinal plants even today. In other words, 72.6 per cent of our present acknowledged medicinal plants can be found in Peter Melius Juhász' Herbarium of 1574. These two figures (99 per cent and 72.6 per cent) probably speak more eloquently than many written pages. The latter were, however, also necessary to account for the figures.

It was left to the end — though it should have been mentioned at the beginning — so as to be given more emphasis that Melius' Herbarium published in 1578 was the first Hungarian book to discuss the rich assortment of medicinal plants in Hungary, and so the first step on the road towards fame the Hungarian medicinal plants have won all over the world.

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AGRICULTURAL PLANTS IN MELIUS' HERBARIUM

Péter Melius Juhász, pastor in Debrecen, published his book "Herbarium. On the names, characteristics and use of trees and grasses" nearly four hundred years ago. It reflected the scientific conception of those days, expressed in all the works that attempted to make the readers acquainted with the utility of plants. A considerable number of these "herbals" summarized the time-tested knowledge of the use of plants. Melius' book did the same in the "Hungarian language"; it was intended to be a guide for the suffering people on their way toward recovery.

On the front page of his book Melius himself emphasized that the useful knowledge on plants had been collected from the "Doctors' books" (completed obviously by his own experiences) and thus made a public property in the Hungarian language. The book presented data on 231 plants, all of which gave the suffering people help in various diseases. It is only natural that the majority of the food plants was omitted from this compilation as they were not directly involved in the cure of the diseases in question. Péter Melius Juhász did not regard the food plants as medicinal plants, although with the condition of the diseased body in view their nutritive effect is no negligible medical aid. Only those plants are included in his book of which he expected therapeutical effects. It is for this reason that only 18 species of the agricultural plants cultivated today are found in the book, hardly 8 per cent of the plants discussed.

The agricultural plants presented by Melius are mostly horticultural plants, first of all vegetables: however, they all agree in containing some (occasionally more than one) active agent which makes them valuable from a therapeutical point of view. Melius emphasized that the plants in question were grown in gardens or in the field, except some species (e.g. clover) which would only become cultivated plants later, though in meadows and pastures they were already useful for the animals.

The vegetables listed in Melius' book and discussed from the point of view of their therapeutical usefulness are the following: purslain *(Portulaca oleracea)*, artichoke *(Cynara scolymus)*, coriander *(Coriandrum sativum)*, poppy *(Papaver somniferum)*, rhubarb *(Rheum rhabarbarum)*, sorrel *(Rumex lapathifolium)*, lettuce *(Lactuca sativa)*, spinach *(Spinacia oleracea)*, asparagus *(Asparagus officinalis)*, horse radish *(Armoracia rusticana)*.

Chicory *(Cichorium intybus)* and ricinus *(Ricinus communis)* — both discussed by the book — only became cultivated plants in Hungary later. These two plants — together with hemp *(Cannabis sativa)* and flax *(Linum usitatissimum)* also mentioned by Melius — have become important industrial plants.

The other plants were not yet cultivated fodder plants at that time, it was only later that they were introduced in production in Hungary. Such are the clovers: red clover *(Trifolium pratense)*, white clover *(Trifolium repens)*, the melilots *(Melilotus officinalis* and *Melilotus albus)*, the fenugreek *(Trigonella foenum-graecum)*. All these plants had to be collected from meadows and pastures if their therapeutical effects were to be utilized.

The agricultural importance of Melius' book lies not only in its listing useful plants and presenting their characteristics, but also in its being a model of how similar works should be published in a popular language.

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FUSARIUM WILT IN WHEAT AND INTEGRATED DISEASE CONTROL IN ROMANIA

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The paper deals with researches and analyses of 80 different wheat cultures in Transylvania and also the results of some factorial and polyfactorial experiments concerning the appearence, behaviour and yield losses caused by fusarium wilt. In 1970 fusariosis was a general epidemic and caused yield losses to 70% in some fields and for Transylvania a 42% average yield loss. The losses varied in relation with the variety and the great variability of the environment and technical conditions. An abundance of humidity, early sowing in September, N. excess, low lodge resistance, a great quantity of weeds, enhanced the appearance and the evolution of the disease, and in consequence determined the high yield losses. The infection on plants during the vegetation also spread on the seeds and this situation should be in the attention of specialists in taking special measures against the diesease preferably as integrated control.

Introduction

Fusarium wilt or ear burn, caused by pathogenic fungi of the *Fusarium* species and particularly the *F. graminearum* Schw. (*Gibberella zeae* Petch), is a widespread wheat disease in Romania (Radulescu, 1957, Hulea 1961); its spread in recent years, may probably be due to the expansion of areas cropped with Bezostaya 1, a wheat variety sensitive to this disease.

The occurrence and epidemic evolution of the wilt under the specific conditions of 1970 in wheat plants infected by *Erysiphae tritici* and *Septoria* species caused high yield losses, ranging from 16 to 70%. In many crops the ears looked as if "burned", necrotic and blackened, and the obtained harvest was of only a few quintals per hectare.

The general occurrence of the disease in all wheat plants, as well as the great variation in damage severity, according to environment and crop science factors, rendered imperative some thorough investigations so as to decide on measures for preventing the disease by integrated control means, without additional expenses. In this paper we present the most important results obtained in our investigations.

Material and Method

So as to establish disease and yield damage extents, 80 wheat crops were investigated, placed on ecologically different soils; mean representative samples of 200—500 plants for each sample were collected from these crops and individually analysed, and the disease degree of the plants was established by categories of severity and a loss corresponding to 50—60 ears for each mean sample.

The correlation between disease and crop science factors was investigated in three factorial experiments laid out in the trial field of the Turda station.

Disease degree was expressed in reaction indices, representing estimates of the product between yield losses by categories of severity and frequency of severity categories, and was written down in figures, one reaction index being conferred to each 10% yield loss.

The figure estimates were statistically computed, using a calculation pattern of simple and factorial variance analysis.

Seed infection degree was established by individual grain analysis in the 1—2 kg representative mean samples during harvesting from the combines in the field or from the storehouses of the agricultural socialist enterprises.

Germination was carried on in the laboratory with germinators on filter paper, while seed was treated with the criptodin preparation at a 100 g/100 kg seed rate.

Results

I. Occurrence, evolution and exhibition of the disease. The disease appears in seedlings during emergence and is transmitted in crops by the infected seeds; it develops depending on the drought conditions directly after emergence, causing variable gaps according to seed infection degree, infection source in the soils, duration and severity of the drought period.

Excessive moisture conditions during spring stimulate the growth of the biological infection storage by propagating the *Fusarium graminearum (Gibberella zeae)* pathogen as saprophyte or as debilitating parasite.

These same conditions stimulate a luxuriant vegetative growth in wheat plants, thus diminishing their resistance to infection.

In the course of the sensitive spring stages, infections occur that first exhibit irregular locuses, where the heading plants grow white in bulks and may be easily distinguished from the green healthy plants.

At this stage the infected plants stop growing and dirty brown stains appear on their organs — roots, stalks, ears — and develop to necroses, while a pinkish — white, violet — rusty mycelium with fungus conidia forms on the tissue surface.

The gradual intensification of the disease process causes a premature drying of the plant that remains partially sterile and with definitely empty grains having a considerably reduced dry weight of only 24.8 g as to 44.3 g absolute weight of the plant with incipient disease.

When perishing, these plants are strongly invaded by saprophyte fungi such as: *Macrosporium* sp., *Alternaria* sp., *Penicillium* sp. etc., that give the plant a dirty grey — blackish colour.

According to plant disease severity, the grain gets differently ill, it sometimes gets empty and shows specific dirty-white stains on the tegument while, in other cases, the plant tegument is partially or totally covered by a felk-like mycelium.

The main damage caused to wheat plants by fusarium wilt is a reduction in number, size and weight of grains by a considerable emptying and a degradation of their seed qualities.

II. Incidence of fusarium wilt and damages caused by it. According to the severity it exhibits in wheat crops, three different categories of diseased plants may be distinguised:

1. Incipiently diseased plants. In the latter the disease evidently appeared during maturity. Disease symptoms are present on the entire plant but only on small portions and particularly on the stalk and palea.

Table 1

Proportion of fusarium affected plants depending on intensity and corresponding crop losses

Degree of disease development	Frequency %		Crop losses %	
	I	II	I	II
Healthy plants	0	0	0	0
First stage of disease	40.3	47.9	10*	10*
Middle stage of disease	44.6	33.9	31.3**	30.8**
Advanced stage of disease	15.1	18.2	68.7**	69.2**

* established by estimation
** established by weighing

All the plants in this category have an internode at the ear base of a normal golden colour, with characteristic dirty dull white stains in between. The discrete presence of a pinkish-white mycelium may be frequently noticed on the palea (Table 1).

2. Plants with moderately severe disease. On these plants the disease occurs at milk stage and causes the complete drying up of the internode at the ear base wich never attains the golden-yellow colour characteristic of a normal physiological maturity but will always have a dull white or dirty white colour going over to brown.

3. Plants with severely developed disease. Plants in this category show developed symptoms that appear at heading — flowering stage and cause a slowing down and stagnation in the normal development of the entire plant and particularly of the ear.

On the ear and on the internode at the ear base advanced characteristic symptoms occur, the tissues get necrotic while on their surface a mycelium and fungus fruiting bodies of a dirty-white pinkish colour appear. The whole plant remains smaller, with a closer ear, with numerous partially or totally sterile spikelets, and their grains are empty and small.

The plants always perish some weeks before normal maturity. Many saprophytes appear between the ear area and its base whose presence convey to the plant its dirty-grey — black characteristic colour. In ratio with symptom severity, the diseased plants incurred specific yield losses, shown in Table 1.

The disease caused higher yield losses in crops on alluvial soils or in those with depressions, in excessively weed infested soils or in soils in which wheat unlodged followed wheat in uninterrupted crops for several years consecutively.

Table 2

Distribution and crop losses caused by wheat fusarium wilt on ecological areas

Area	Area designation	Frequency of diseased plants depeding on disease severity				Crop losses, estimated, %
		S	I	M	E	
I	The Transylvanian Plain the Cluj, Dej and Sălaj hills	0	39.7	49.3	11.0	26.84
II	Medium course of the Mureș river	0	13.6	52.9	33.5	40.87
III	Upper course of the Mureș river, the lakes region — Luduș-Zaul de Cîmpie	0	1.7	27.9	70.3	57.35
IV	Confluence watershed of the Mureș and Sebeș river	0	6.4	52.5	41.1	45.27
	Average	0	15.3	45.6	39.0	42.58

S = healthy plants
I = first stage of disease
M = middle stage of disease
E = advanced stage of disease

In these crops, existent in the third and in fourth zone (Table 2), highest losses registered reached 67% and lowest 28%, while in the second zone highest losses reached 59% and lowest 32%.

Most of the ears in these crops were "burned", partially or totally sterile, necrotic and blackened, with excessively empty grains and yields did not exceed several quintals per hectare.

Yield losses presented a 40.87% mean in the second zone, a 57.35% mean in the third zone and a 45.27% mean in the fourth zone.

In crops situated on permeable, weed-free soils the disease appeared with moderate severity, the number of plants incipiently and moderately diseased predominated and mean yield losses were estimated at 26.48% (first zone).

The data obtained show the unprecedented, particularly damaging character by which fusarium wilt manifested itself in wheat in 1970, mean losses in some areas cropped to wheat under conditions prevailing in the Transylvanian Plain being estimated to 42.58%.

III. Effect of environment and technique on fusarium wilt. Occurrence and severity of fusarium wilt in wheat is in strong connection with a series of environmental conditions and technical factors.

The disease appears with more severity during the rainy years on the soils with moisture excess and under conditions of high air moisture. Under moist conditions, pathogen propagation and spreading occurs more rapidly, and the plants, debilitated through water lodging oppose only reduced resistance to the disease.

1. As may be seen in Table 3, severe weed infestation also favours occurrence and severity of fusarium wilt, as by competing plant development the weeds render the plants more sensitive and create a microclimate favourable to successive infections.

2. Under conditions of fusarium wilt epidemic occurrence plant resistance to lodging is an extremely important factor. The lodged plants get more severely ill, they cannot rise up even partially and this aggravates yield reduction. It may be seen from Table 3 that in a lodged and weed infested field a 6.5 reaction index is obtained and a mere 812 kg/ha yield, while in a normal crop the yield is of 2.639 kg/ha grains, and the reaction index is of only 3.4.

3. The previous crop greatly influences plant disease and yield loss due to the biological storage of the pathogen that varies according to the species of crop the year before.

Table 3

Influence of weed infestation rate, lodging resistance and preceding plants on fusarium wilt in wheat

No.	Items — classifications	Average reaction index		Diff.	Signif.	Yield kg/ha
		$\bar{x}$	%			
	I. Crop condition					
1	Wheat overgrown with weeds	5.3	155.9	1.9	+++	1214
2	Wheat in normal cultivating condition	3.4	100.0	0	mt.	2803
	II. Lodging resistance					
3	Lodged and weedy wheat	6.5	191.2	3.1	+++	812
4	Wheat with normal plants position	3.4	100.0	0	mt.	2639
	III. Preceding plants					
5	Wheat after weedy wheat	5.5	204.7	2.8	+++	1109
6	Wheat after bean	3.0	111.1	0.3	—	3182
7	Wheat after corn	2.7	100.0	0	mt.	3102

Thus, in wheat grown after wheat, the disease reaction index was 5.5, as to only 3.0 and 2.7 for wheat grown after beans, and yield was of only 1.109 kg/ha, as to 3.182 and 3.102 kg/ha in wheat grown after beans or after corn respectively — our data in this analysis correspond with those reported by DICKSON in (1956).

4. Fertilization greatly affects plant disease severity. The lowest disease severity of 1.7 reaction index was found in plants grown in soils with an $N_{40}P_{60}$ fertilizer rate.

When phosphorus was given in a P_{60} rate the reaction index of disease severity increased up to 1.8 the difference from the control was not significant but on such a soil yield decreased by 602 kg/ha as to the control.

As the nitrogen rate increases to N_{80}, N_{120} and N_{160} while the phosphorus rate is constantly kept at P_{60}, the disease index increases to 2.3, 3.0 and 3.8 respectively, the differences of disease degree from the control as well as the differences between the differently fertilized soils being highly significant. Yields at P_{60} N_{80} may be considered practically equal to those obtained in the control, but in the other fertilized soils yields decreased with 351 kg/ha and 508 kg/ha respectively as a consequence of the high increase of disease severity (Table 4).

Table 4

Influence of basal dressing on fusarium wilt in several varieties and strains of wheat

No.	Items — classifications	Average reaction index on basal dressing		Diff.	Signif.	Yield kg/ha
		$\bar{x}$	%			
	I. Variety					
1	Bezostaya	2.9	181.3	1.3	+++	4009
2	Harrach	2.8	175.0	1.2	+++	3771
3	T 194	2.7	168.8	1.1	+++	4347
4	Dacia	2.5	156.3	0.9	+++	4027
5	Favorit	2.7	150.0	0.8	+++	3967
6	T 195	1.6	100.0	0	mt.	4414
	DL 5%			0.34		
	II. Basal dressing					
1	$N_{160}P_{60}$	3.8	223.5	2.1	+++	3870
2	$N_{120}P_{60}$	3.0	176.5	1.3	+++	4027
3	$N_{80}P_{60}$	2.3	135.3	0.6	+++	4395
4	$N_{0}P_{60}$	1.8	105.9	0.1	—	3776
5	$N_{40}P_{60}$	1.7	100.0	0	mt.	4378
	DL 5%			0.13		

From the data it appears that fertilizers increase plant susceptibility to fusarium wilt when an optimum rate between N and P is not observed, and the disease degree gets higher as the nitrogen rate increases (Table 4).

5. Interaction between fertilizer × variety in different varieties. Chemical fertilizer activity specifically affects disease degree according to N and P ratio and in correlation with variety resistance (Table 5).

The Bezostaya variety proves a constant variety sensitive to all 5 previously fertilized soils. This variety apparently reacts by relatively reduced differences in disease severity as dependent on nitrogen increase.

Table 5

Reaction to fusarium wilt in basal dressing × variety interaction

Basal dressing	Variety											
	Bezostaya		Harrach		Dacia		Favorit		T. 194		T. 195	
	Reaction index	S	Reaction index	S	Reaction index	S	Reaction index	S	Reaction index	S	Reaction index	S
N_0P_{60}	2.9	—	1.3	—	1.4	+	1.9	—	2.1	+	1.1	—
$N_{40}P_{60}$ mt.	2.6	—	1.1	—	1.8	—	2.2	—	1.6	—	0.8	—
$N_{80}P_{60}$	3.2	+	3.6	+++	1.9	—	2.1	—	1.8	—	0.9	—
$N_{130}P_{60}$	2.9	—	3.8	+++	3.4	+++	2.4	—	3.8	+++	1.4	+
$N_{160}P_{60}$	3.1	+	4.2	+++	4.0	+++	3.6	+++	4.2	+++	3.8	+++
DL 5%	0.4		0.4		0.4		0.4		0.4		0.4	

In all the other varieties: Harrach, Dacia, Favorit as well as in the T 194 and T 195 lines a resistance reaction was emphasized. Conditioned by an adequate fertilizer balance these varieties react as resistant to disease in a well balanced fertilization and some of them even at higher N rates, but an increase of nitrogen to N_{80} for the Harrach variety, to N_{120} for the Dacia variety and the T 194 line and to N_{160} for the Favorit variety and the T 195 line brought about a strong sensitivity in these varieties and a disease degree more severe than in the sensitive Bezostaya type with highly significant differences (Table 5). The severe disease degree in excessive nitrogen rates was also directly influenced by plant lodging, according to their specific requirements.

The data prove that an excess in nitrogen increases plant susceptibility to fusarium wilt; the rates at which nitrogen is used and the ratio between N and P are characteristic for each cropped variety individually and fertilization may be considered as one of the most important and economically efficient technical means of preventing the disease and of reducing yield losses.

6. Planting time, planting stand and maintenance practices clearly influence the disease.

In a factorial experiment carried out on the Bezostaya variety grown after corn, a stronger attack was registered when wheat was planted on September 15th than when planted on October 5th and 25th (Table 6). The effect of planting time should also be analysed keeping in view conditions characteristic for autumn droughty periods that favour seedling primary infection with Fusarium sp.

Table 6

Influence of cultural practices, density and sowing time on fusarium wilt in wheat

No.	Items — classifications	Average reaction index		Diff.	Signif.	Yield quintal/ha	Weeds number
		$\bar{x}$	%				
	I. *Cultural practices*						
1	Uncultivated, nutrition applied	2.86	112.5	0.32	+++	3191	186.1
2	Treated with Igran 5 l/h seeder	2.54	100.0	0	mt.	3216	111.0
3	Cultivated with rotary hoe	2.53	99.6	−0.01	—	3129	135.0
	DL 5%			0.11			
	II. *Sowing density*						
1	200 wg[1]/mp	2.49	87.3	−0.36	000	2916	—
2	400 wg/mp	2.85	100.0	0	mt.	3286	
3	600 wg/mp	2.60	91.2	−0.25	000	3332	
	DL 5%			0.07			
	III. *Sowing time*						
1	Sept. 15	3.39	140.7	0.98	+++	2920	—
2	Oct. 5	2.41	100.0	0	mt.	3319	—
3	Oct. 25	2.13	88.3	−0.28	000	3297	
	DL 5%			0.13			

[1] wg = wheat grain

Maintenance cultural practices also show their influence on disease severity, and in the first place concerning weed infestation degree (Table 6).

7. Variety reaction to fusarium wilt. Under conditions prevailing in 1970 none of the cropped wheat varieties proved immune.

Most varieties exhibited a high susceptibility degree and registered yield losses ranging between 30 and 50%.

A more reduced number of varieties proved resistant and registered yield losses ranging between 18—26% (Table 7).

The Avon, Neuzucht, Monon, Poncheau and other varieties showed a specific resistance during stages up to grain formation, but up to full matu-

Table 7

Reaction of some releasive wheat varieties and parents to fusarium wilt

No.	Variety	Rate of diseased plants in grain formation stage %	Reaction index %	Reaction index %
1	Cenad 117	31.9	5.9	327.7
2	București 1	22.0	5.9	327.7
3	Avon	5.4	5.7	316.6
4	Varietas 186	29.9	5.1	283.3
5	Ponca	35.9	5.0	277.8
6	Odesskaya 16	40.2	4.9	272.2
7	Neuzucht	1.7	4.7	261.1
8	Nr. 301	42.0	4.5	250.0
9	Odvoș 241	12.6	4.4	244.4
10	Cluj 11/54	11.5	4.3	238.8
11	San Pastore	43.1	4.0	222.2
12	Vermillon	34.7	4.0	222.2
13	Bezostaya 1	48.4	3.7	205.5
14	Monon	0	3.6	200.0
15	Skorospelka 3B	34.2	3.6	200.0
16	Rudest	6.8	3.6	200.0
17	Poncheau	0.9	3.6	200.0
18	Concho	54.6	3.4	188.9
19	Cluj 722	31.5	3.4	188.9
20	Triumph	61.8	3.3	183.3
21	Endress st. 451	2.2	3.2	177.8
22	Rannaya 12	31.3	3.1	172.2
23	Cappelle Desprez	0.9	3.1	172.2
24	Gaines	7.0	3.0	166.7
25	Maître Pierre	5.1	3.0	166.7
26	Mitchurinka	3.9	2.6	144.4
27	Harrach	0.9	2.5	138.8
28	Hibrid 5407	0	2.5	138.8
29	Favorit	17.2	2.5	138.8
30	Camplein	18.0	2.4	133.3
31	Lutescens 17	4.0	2.4	133.3
32	Etoile de Choisy	6.6	2.0	111.1
33	Belotserkovskaya 198 mt.	2.5	1.8	100.0

rity they lost their resistance exhibiting a high susceptibility degree, characterized by reaction indices ranging from 3—6 to, 7.

A high tolerance degree was exhibited by the Triumph variety although when recording disease appearance Triumph proved the most susceptible in all soils with a 61.8% rate of diseased plants, but up to full maturity this variety tolerated the disease well, and achieved a mean reaction index of only 3.3 (Table 7).

Table 8

Rate of wheat grain infection with fusarium sp. of Bezostaya variety in some wheat crops

Sampled crops	Normal grains which are apparently healthy %	Diseased grains	
		Shrivelled with symptoms %	With mycelium %
1	95.1	4.1	0.8
2	92.0	7.2	0.8
3	91.5	8.0	0.5
4	90.7	6.7	2.6
5	89.2	8.2	2.6
6	87.0	11.3	1.7
7	86.6	12.5	0.9
8	85.6	13.6	0.8
9	84.8	14.5	0.7
10	83.0	17.0	0
11	80.4	18.1	1.5
12	79.8	18.0	2.2
13	79.6	13.3	7.1
14	79.4	18.5	2.1
15	78.3	20.2	1.5
16	75.5	20.2	4.3
17	73.2	24.2	2.6
18	71.2	28.8	0
19	69.7	27.1	3.2
20	63.8	35.4	0.8
21	57.5	42.5	0
22	54.0	43.8	2.2
23	51.8	47.5	0.7
24	47.0	52.3	0.7
Average	77.0	21.4	1.6

Table 9

Infection rate of wheat grains with fusarium sp. in some releasive long term varieties

No.	Variety	Normal grains which are apparently healthy	Diseased grains	
			Shrivelled with symptoms	With mycelium
1	Favorit	87.0	13.0	0
2	Caucaz	82.4	17.6	0
3	Jubileinaya 50	80.8	19.2	0
4	Ponca	80.4	19.4	0.2
5	Aurora	77.8	22.2	0
6	Rannaya 12	74.0	26.0	0
7	Excelsior	72.5	27.0	0.5
8	Harrach	69.4	30.6	0
9	Skorospelka 3 B	65.8	33.5	0.7
10	Dacia	64.4	30.6	5.0
11	Bezostaya 1	63.1	35.2	1.7
12	Moldova	60.3	37.2	2.5
13	București 1	49.6	48.1	2.3
	Average	71.3	27.7	1.0

The most sensitive varieties were: Cenad 117, București 1, Avon, Ponca, Bezostaya 1 and others, while the Belotserkovskaya 196 Etoile de Choisy, Lutescens 17, Favorit and Harrach varieties exhibited a greater resistance.

IV. Fusarium wilt infection in seeds. From plants with *Fusarium* wilt the infection moves to the seeds already during the vegetation period, and wheat grains get a different disease degree according to grain infection time, to disease severity in the plant and to moisture microclimatic conditions occurring at ear level.

Generally infection incidence in seeds is much lower than that in plants, as the grains are protected by paleas and have a specific organo-trophic resistance.

From the analysis of a number of 24 wheat samples of the 1970 harvest of Bezostaya variety, originating from 24 localities it appears that 77.0% of the grains are apparently healthy on average and 23% of the grains are evidently ill, but severity degrees are different (Table 8).

Mean percentage of apparently healthy grains in 13 varieties was of 71.3%, mean percentage of empty grains with symptoms was of 27.7%, and mean percentage of diseased grains with evident mycelium was of 1% (Table 9).

Under germination conditions, the healthy apparently normal grains germinate in a proportion of only 77% and 23% of the seeds with symptoms do not germinate or germinate inadequately, in a percentage of only 52%.

This is why a seed conditioning by selection forms an efficient means of improving plant health and cultural value of the seed.

In the selected seed the percentage of normal, apparently healthy grains increased on average in 9 samples from 77.2% to 85.9%, empty grain percentage with symptoms considerably decreased from 21.8 to 13.4%, those with evident mycelium from 1% to 0.7% and their germination increased by 8.3% as compared to the germination of non-selected seeds.

Seed treatment with criptodin reduced the percentage of diseased grains and provided a germination increase in the sample of 11.6% as to the non-selected and non-treated seed.

Discussion

Fusarium wilt epidemic in wheat of Transylvania appeared earlier than in the previous years and occurred on plants already severely infected with *Erysiphae tritici* and *Septoria* sp. prior to *Fusarium;* it developed relatively acutely and manifested itself in the damage form never encountered in our country.

The disease appeared everywhere on all wheat plants under cultivation; it however caused varied yield losses essentially depending on the moisture excess specific for 1970 and on the diversity of environmental and technical factors specific to wheat crops. From the investigations it appears that fusarium wilt had a general, epidemic spreading and caused high damages on all wheat planted areas in our country; in the Transylvania Plain mean losses were estimated at 42%. Highest yield losses were registered in moist alluvial soils, in weed infested soils and in those planted early in September. In these cases the areas sown with wheat incurred yield losses of up to 70%, and yields hardly reached 3—6 q/ha.

Smaller losses were registered in wheat crops sited on permeable soils of the hilly slopes with normal rim-off, specific to the Transylvania Plain, free of weeds and well aired. In such occurrences yield damages for 1971 were minimal, ranging from 16.8 to 25%.

The main damage effect of fusarium wilt consisted of a decrease in seed size and weight, as well as in the shrivelling and infection of the grains leading to a depreciation of their qualities.

The study of the disease development and damages evidence a strong relationship between the destructive action of the disease as function of cultural practices and the natural environment.

The premises for the appearance of certain disease focuses with the plants exposed to infections are increased by early autumn sowing, during September, of an infected seed, unselected and untreated, on a previously infected land.

The disease exhibits a progressive development on lands with excessive moisture and severe weed infestation; these conditions are favourable both for repeated, generalized, infections and for intensification of the destructive action of the disease.

The same ecological conditions of excessive soil moisture and severe weed infestation diminish the plant resistance to pathogen.

On the contrary, sowing during the optimum period as well as application on the square meter of an optimal quantity of seed with high germinability, on a soil fertilized in equilibrated levels and rates, without any N-excess, accompanied by the utilization of a healthy, selected and treated seed on soils with high permeability and after row plants or legumes as previous crops, avoids the important primary infection sources and gives the possibility to obtain closed stands exhibiting a good healthy state.

Avoiding the lands with severe weed infestation as well as rigorous weed control contribute to a better soil aeration under the crops and creates unfavourable conditions for the infections with *Fusarium* sp. The crops are not further weakened by the weeds and consequently the yield losses are reduced.

The resistance of different wheat varieties to fusarium wilt in connection especially with N excess and lodging resistance may be rendered profitable only by avoiding excessive N fertilization and by keeping strictly to the optimum rate between the nitrous and phosphatic fertilizers.

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FLUORESCENCE SPECTRA OF ACTIN

By

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In our experiments we studied the excitation and fluorescence spectra of actin, and the latter's changes. The excitation and fluorescence spectra of actin were measured with spectrofluorometry. The excitation spectra of actin were found to change to a higher, while the fluorescence spectra to a lower extent. While purifying the actin we found that the spectra showing a more homogeneous, narrower band become wider—(290—310 nm) contradicting the process of purification. Although purification, gel-filtration, ultracentrifuging, polymerization and depolymerization, dialysis result in G-actin as proved by the structures of the ultra-violet spectrum and the difference extinction spectrum, an increase in the polymerization ability, and even by the substances removed, the broadening of the excitation spectrum suggests secondary, tertiary structural changes, disintegration rather than stabilization. Broadening of the spectrum indicates a relationship between the loosely and closely bound lipid components removed from the protein. We have produced the lipid components of gel-filtrated actin and present its excitation and fluorescence spectra as well as its thin-layer chromatogram.

Introduction

We have observed for some time that actin has a hypochrome effect, the extinction value per 1 mg protein as measured in the ultra-violet range increases upon standing. Recently we have also noticed that a temporal increase in the value of E_{280} occurs in gel-filtrated actin too, which can be explained only by changes in the secondary and tertiary structures of the molecules. The latter seems all the more probable as the value of E_{280} increases in non-gelfiltrated actin even after Dowex II and Dowex 50 treatments which remove the peptids and inoic fractions. When dialysed against distilled water low molecular weight substances appear in the dialysing water and their release can be followed up in time. Beside the ultra-violet spectrum the changes of the excitation and fluorescence spectra also provide a possibility to study the actin structure.

In our present study we deal with actins obtained in the different phases of actin purification, and the excitation and fluorescence spectra of gel-filtrated actin which is considered to be the purest — as it consists of a single polypeptid chain. From the results we wish to draw conclusions on the structural stability of actin molecules. Not long ago we reported (Fazekas *et al.* 1968a) that myosin ATP-ase — when the myosin is prepared from a muscle grinding with Szent-Györgyi's (Szent-Györgyi 1949) method modified by Portzehl *et al.* (1950), as well as myosin ATP-ase prepared from isolated myofibrillum according to Perry—Gray (1956) are best activated by non-purified crude actin and show a lower ATP-ase activity with gel-filtrated F-actin, in spite of the fact that purified gel-filtrated actin has the highest polymerizing ability. We obtained the same result with myosin chromatographed previously on a DEAE-cellulose column. We also reported (Fazekas *et al.* 1968b) that the gel-filtrated actin showed more components — including low molecular weight substances — when repeatedly gel-filtrated. Furthermore, at the Prague Congress we pointed out (Fazekas—Székessy—Hermann 1968c) — that from purified actin 8—9000 g lipid/60000 g actin could be removed with a lipid solvent after which the actin lost its water solubility as well as its myosin activating- and polymerizing ability.

Material and Method

The actin was produced from muscles of not more than four months old rabbits according to Szent-Györgyi's method (Szent-Györgyi 1949) with the modification that the residue obtained after the extraction of myosin was washed with solutions of 0—4°C until the acetone dried powder was produced. The actin was extracted from the dried acetone powder with a solution of 2.5×10^{-4}M ATP, 2×10^{-4}M 2-mercaptoethanol (pH 7.2) (20 ml/g) cooled to 0°C. Henceforth we followed the method used by Rees—Young (1967) with the difference that before gel-filtration the materials charged with ions were removed with Dowex 50 and Dowex II (from Fluka AG) treatments according to Drabikowski—Pisarek (1964). Gel-filtration was carried out on a 2.3×52 cm Sephadex G* 200 column with maximum 10—12 ml (4—5 mg/ml) actin solution applied. With this standardized column used for several years now the actin obtained always gives similar results eluating at a value of 0.82 KD. In the examination the peak tubes of the well polymerizing actin was used.** Fractions of 6.2 ml were collected.

The Ca-ion was left out of the extracting solution, since actins extracted without Ca-ions were found to be more homogeneous.

The protein content of the fractions was calculated according to the Kjeldahl N-determination. The N content of the fractions was taken for 16 per cent except the gel-filtrated fraction in which the N content was taken for 16.1 per cent. In addition, the concentration of gel-filtrated actin was also calculated on the basis of UV absorption (on Beckman spectrophotometer DU model G 2400), since on the basis of the Kjeldahl value the absorption of 1 mg protein had been found to be 1.125 E_{280}, at pH 7.0, while 1.095 E_{290} at pH 13.0. The values obtained with a basic pH can be used for F-actin and depolymerized actin as well, with a possible error of 3—5 per cent.

5—10 ml of the gel-filtrated actin was kept in a refrigerator for 4—6 hours, till the beginning of the spectrofluorometric test, while the larger part was dialysed against distilled water until it was completely ion-free and, after sampling the lipid was extracted.

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* Tyr, tyrosine; Trp, tryptophan; Tris, tris (hydroxymethyl amino-methane; and ATP, the 5' (pyro)-triphosphates of adenosine; from Reanal, Budapest, Hungary; Sephadex G-200, from Pharmacia, Uppsala, Sweden; 2-mercaptomethanol, from Schuchardt, Munich; k_D, equilibrium constant; ηs_p = specific viscosity = $(\eta/\eta_0) - 1)$; (η/η_0) = relative viscosity.

** The term "well polymerizing actin" means that in a 2 ml capacity Ostwald viscosimeter of 30 sec flow time, at 24°C it reaches or exceeds the value of ηsp = 0.63 within seven minutes.

The lipids were extracted from the dialysed actin by Folch's method (Folch *et al.* 1957). Extraction was carried out with 20 volumes of a mixture of chloroform: methanol (2 : 1, v/v) and the extract washed in a N_{φ} atmosphere with a mixture of 0.1M KCl : methanol : chloroform (94 : 96 : 6, v/v/v). The lipids were dried in a rotary evaporator and resolved in N_{φ} enriched chloroform. They were saturated with N_{φ} gas and filled up to a volume of 10 ml. From completely dialysed actin a varying amount of lipids averaging 11.5 per cent, while from non-dialysed, gel-filtrated actin even more were obtained, when they were calculated after the gravimetric determination.

Fluorescence methods. Fluorescence measurements were made with a Hitachi—Perkin—Elmer MPF 2/A spectrofluorimeter and the conventional optical system for the detection of fluorescence at 90° relative to the path of excitation light. A quarz cuvette of 1 cm length was used. The samples were excited by an irradiation of 280 nm wavelength except crude G-actin which was irradiated with a wavelength of 290 nm (Fig. 3). Diffuse light was filtered at 290 nm by means of a cut-off filter except crude G-actin in which filtering took place at 310 nm.

A Xenon lamp of 150 W was used as a light source.

Results

Fig. 1 shows actin fractions obtained by gel-filtration.

Fraction III is the only fraction that polymerizes. Fraction IV is of low molecular weight and can be removed by dialysis. For the purpose of our investigations only the peak tubes of fraction III were used.

The excitation and fluorescence emission spectra were determined for each actin fraction obtained in the course of purification. The fluorescence spectrum was measured as excited by a monochromatic radiation of 280 nm wavelength. The excitation and fluorescence emission maxima of the fractions are summed up in Table 1.

Figures 2—6 show the excitation and fluorescence spectra of the actin fractions. Fig. 2 shows actin extracted from the acetone powder, without puri-

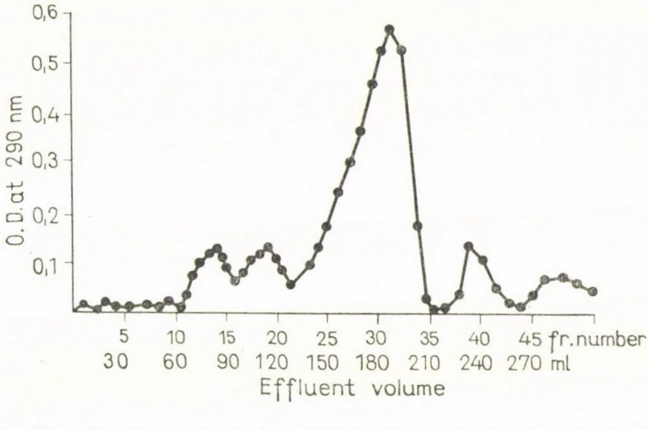

Fig. 1. Gel-filtration of actin on Sephadex G-200 column (2.3 × 52 cm). Actin was extracted from acetone-dried powder with 2.5×10^{-4}M ATP and 2.5×10^{-4}M 2-mercapto ethanol. The column was equilibrated and eluted with the extracting solution. Effluent volume, 6.2 ml per fraction

Table 1

Maxima in excitation and fluorescence spectra of actins

	Excitation max. nm	Emitted fluorescence max. nm
Crude actin	298	355
Actin-F mg/ml	292	335
Gel-filtrated actin-G	293	337
Gel-filtrated dialysed actin-G	288	340
Actin-F at 3.8 mg/ml conc.	298	329
Lipid fraction	340–350	360, 373, 407

fication. Fig. 3 shows the excitation and fluorescence spectra of F-actin purified in a Spinco Ultracentrifuge (Model L.) at 105000 g, over two hours, which was then resolved — while stirred — in 0.1 M KCl, and the rough parts removed by centrifuging at 2000 g. Fig. 4 shows the excitation and fluorescence spectra of F-actin at a higher concentration (3.8 mg/ml). In Fig. 5 the excitation and fluorescence spectra of gel-filtrated actin in the eluting solution, while in Fig. 6 the excitation and fluorescence spectra of the same actin after complete dialysis can be seen. Fig. 7 shows the excitation and fluorescence spectra of lipids obtained from dialysed actin in chloroform. In Fig. 8 the lipid fractions of gel-filtrated actin are shown after having been separated with thin-layer chromatography on a silica gel plate.

When the figures are compared the most conspicuous phenomenon is that the excitation band of the actin gradually widens from the crude to the

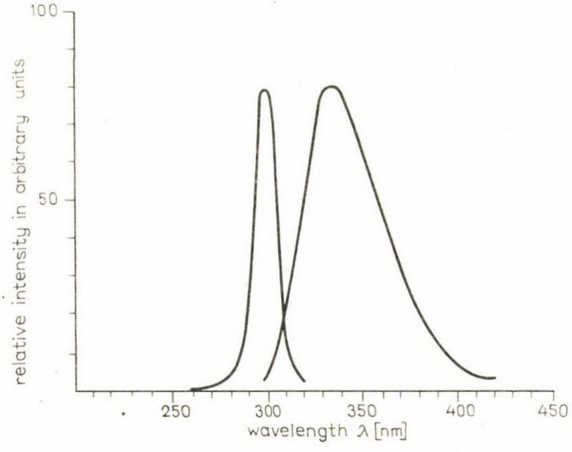

Fig. 2. Excitation and fluorescence spectra of crude actin-G extracted from acetone-dried powder. Protein concentration: 1 mg/ml

gel-filtrated actin. While the "effective band width"* of crude, well polymerizing actin falls almost completely between 290 and 310 nm (beginning with a very low effect and practically ceasing at 320 nm), and dominates the greater part of the spectrum, the "effective band width" of gel-filtrated G-actin falls between 270 and 310 nm with the lower limit of the excitation spectrum being at 240 nm and the upper limit remaining unchanged. The activating spectrum further broadens with the dialysed G-actin, where the "effective band width" falls between 260 and 310 nm, and the lower limit of excitation

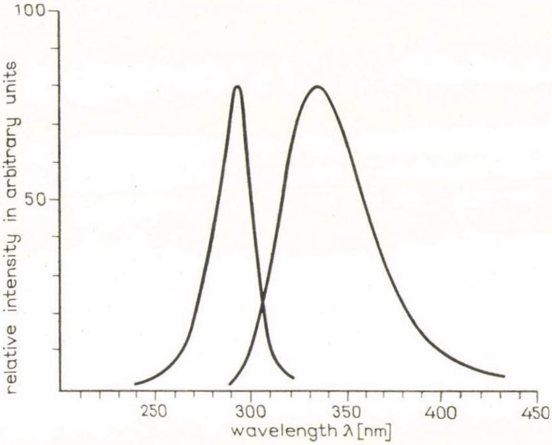

Fig. 3. Excitation and fluorescence spectra of centrifuged and resolved actin-F at 1 mg/ml concentration. Actin-F was resolved in 0.1 M KCl

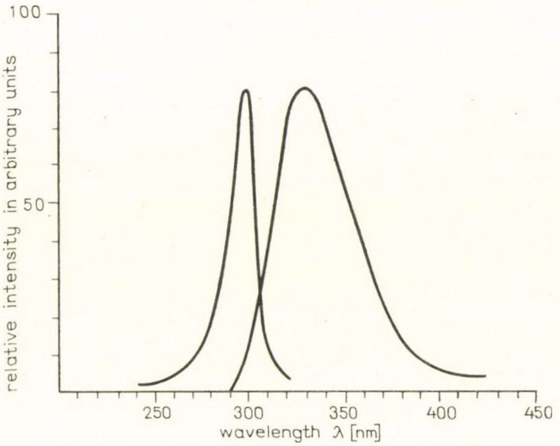

Fig. 4. Excitation and fluorescence spectra of actin-F at 3.8 mg/ml concentration

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* The area contains the greatest part of the activating spectra.

begins at an even shorter wavelength, 230 nm, while the upper limit is shifted beyond 340 nm. Taking into consideration that not more than 20 per cent of the proteins are lost in the process of purification — a part of which is actin —, and other losses are not of protein origin, there is a contradiction between the process of purification and the structure of the excitation spectra. Parallel with the substances removed it is the steadier, more homogeneous state of the narrower band widths that should be reflected in the spectra. This contradiction can only be explained by structural changes occurring in the molecule during purification.

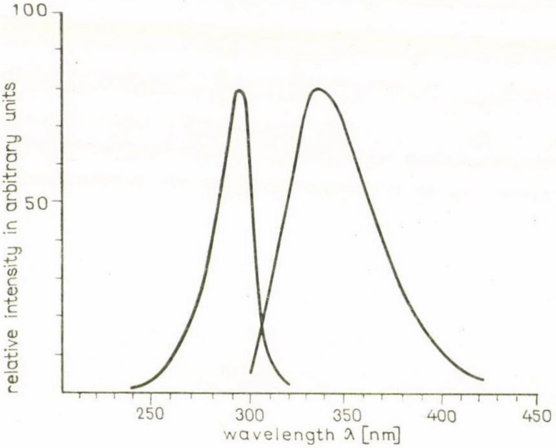

Fig. 5. Excitation and fluorescence spectra of gel-filtrated actin at 1 mg/ml concentration

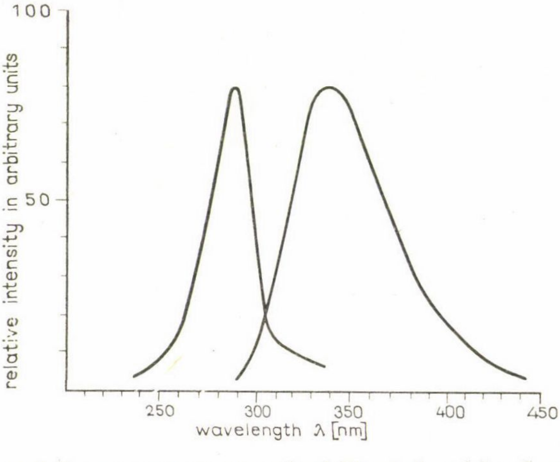

Fig. 6. Excitation and fluorescence spectra of gel-filtrated and ion-free actin-G at 1 mg/ml concentration

On the other hand, in the course of actin purification the ion exchangers (like the Dowex-es) also remove mainly lipids, nucleotids, ions and an insignificant amount of protein. Furthermore, fraction I obtained by gel-filtration on Sephadex G-200 column also contains mostly lipids, first of all phospholipid.

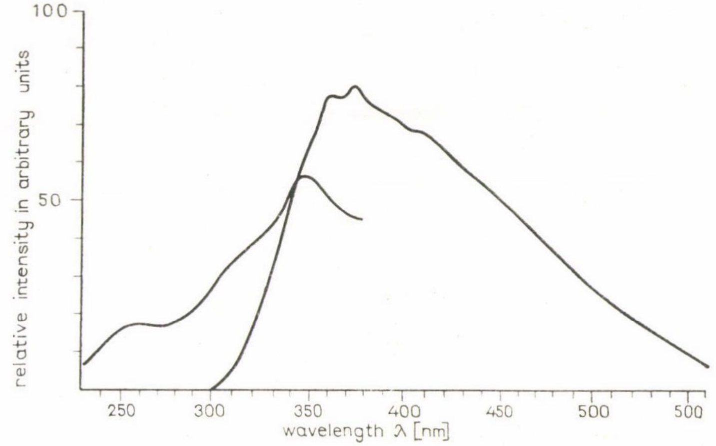

Fig. 7. Excitation and fluorescence spectra of isolated lipid in chloroform

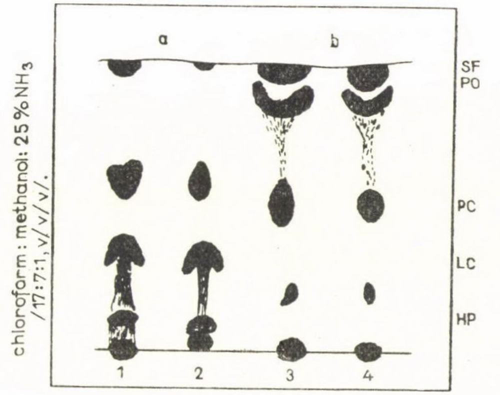

Fig. 8. Thin-layer chromatography of lipids from gel-filtrated actin by Cuzner-Davison, J. Chromat., *37*, 388 (1966): a) 1,2 lipids from dialysed actin; b) 3,4 the same after peroxidation. (HP Hydroperoxid, LC Lysophosphatidyl cholin, PC Phosphatidyl cholin, PO Peroxides, SF Solvent front)

The gel-filtrated actin contains 10—11 per cent lipid as well. This lipid — as shown by Fig. 8 — is not homogeneous. At room temperature freshly isolated lipids are mixtures of solid and liquid components, while in the open air the liquid part sooner or later becomes intensely yellow then red-brown as a consequence of peroxidation and aspecific polymerization. In such cases two more definite spots appear in the chromatogram, one of them near the starting line, while the other, the red-brown spot, along the front. Freshly isolated lipids always have a characteristic smell which disappears after a few hours, but before the discoloration.

Polymerization is the best in gel-filtrated actin, while in the case of actin dialysed against distilled water it becomes prolongated and reaches a maximum only between the 15th and 20th minute (instead of 7 minutes), and even then only a value of η sp 0.40—0.50 instead of 0.3. By concentrating the dialysing water and with the aid of lipid solvents a varying amount of lipid can be extracted from the residue, which confirms the theory that it is lipids that maintain the polymerizing ability. Referring to what was said in the introduction, namely, that the ATP-ase activity of myosin is best promoted by crude actin rather than by purified F-actin we may assume that in the myofibrillar structure the integrity of actin must be ensured by a still greater amount of lipid.

During the purification of the actin the lipids bound loosely on the surface of the molecule and more closely in its structure are gradually removed, and the coverage of excitable Tyr and Try parallelly decreased. When the coverage of buried Tyr and Try has decreased a loosening of the structure of the molecule becomes possible, as is indicated by the broadening of the excitation band. The broadening of the excitation band is, in turn, the measure of the disintegration in the molecule. All that has been told is well supported by Cowgill's works. Cowgill (1968a, 1968b and 1968c) points out that when the detergents remove the lipids from the proteins the width of the excitation spectrum and the fluorescence value originating from Tyr measured at 305 mn increase. The essential lipid content of actin has not yet been determined with exact numerical data. On the basis of the data (4—5 mg lipid/45 mg protein) the ratio of molecule lipid/molecule actin can be reckoned — depending on the quality of the lipids. Owing to the absence of other information any further conclusion is, however, considered unreasonable and will therefore be avoided.

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ANATOMICAL, ULTRASTRUCTURAL AND PHYSIOLOGICAL STUDIES ON THE PRIMARY CORTEX OF EUONYMUS EUROPAEUS L. DISPLAYING PHOTOSYNTHETIC ACTIVITY II.

SEASONAL CHANGES

By

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The authors studied the seasonal ultrastructural and physiological changes of chloroplasts in the chlorenchyma of one- and two-year-old shoots of *Euonymus europaeus* L. by means of electron microscopic examinations and $^{14}CO_2$ incorporation. The reactions of the plasts to the same stimulus at a given time were not quite identical, but in essentials, they maintained their basic inner structure required for their activity throughout the year. In the lamellar structure the authors found qualitative, and — regarding the number of grana — even quantitative seasonal changes. The number of grana considerably decreased by October and remained at a low level even in January; in March and April it increased. The seasonal course of the total amount of chlorophyll was parallel with this. The results of the studies in these two directions confirm one another. The annual course of photosynthetic activity can be parallelled with the phenology of the plants. At the end of winter and at the beginning of April, the photosynthetic activity is very high. At the beginning of April the synthesis of high molecular weight substances may have a role in the sprouting of the bud. At the time of leaf function the activity of the bark decreases, biosynthesis shifts toward forming directly available low molecular weight substances.

Introduction

In a previously published paper (Szujkó-Lacza—Rakován—Fekete—Faludi-Dániel 1971) the authors gave an account on the anatomy, ultrastructure, chlorophyll content and photosynthetic activity of tissues with green chloroplasts in the shoot axis of *Euonymus europaeus* possessing fully developed foliage. From their observations the authors drew the conclusion that these chloroplasts — especially those in the outer cell-layers of the cortex — are able to photosynthesize. In order to be able to draw conclusions on the possible role of these tissues in the life of the whole plant studies were required on their seasonal changes. The authors first approached the problem from a cyto-ecological aspect, i.e. they wished to get an answer to the question: what structural changes occur during the year in the plastids of the bark — green even in winter — of this deciduous species, and what influence do these structural changes have on the photosynthetizing ability of the plastids.

There are data available on the seasonal changes of photosynthesis. It is known, for example, that leaves of conifers and evergreen broad-leaved species are able to display in vivo photosynthetic activity in winter, at temperatures below freezing-point (cf. STÅLFELT 1960, UNGERSON—SCHREDIN 1965). There is no evidence, however, of there ever having been any special studies on the seasonal changes of photosynthesis in the axial chlorenchyma.

Literature on the structure of the chloroplasts in organs green in winter contains highly different results. KRASAVTZEV—TUTKEVICH (1971) when examining the cortical parenchyma of *Betula* and *Sambucus* found differences in the lamellae of summer and winter plastids. On the other hand PARKER—PHILPOTT (1963) found no fundamental structural differences between winter and summer chloroplasts in *Pinus strobus*- and *Rhododendron* leaves. For literature on the seasonal changes in chlorophyll content and on light-microscopic studies related to the subject see SZUJKÓ-LACZA—FEKETE—FALUDI-DÁNIEL 1970, SZUJKÓ-LACZA *et al.* 1971.

A better knowledge of seasonal changes in the chlorenchyma of the shoot axes may be acquired by the joint study of function and structure. This is the aim the authors had in mind in this work.

Material and Method

For the purposes of seasonal examinations samples were taken between July 1970 and April 1971, at phenologically characteristic stages of *Euonymus europaeus:* in July when the foliage had fully developed; in October when the foliage had turned yellow; in January, in the period of perfect rest; in February–March when water circulation was starting and the buds were beginning to swell; and in April and May when the young foliage was developing.

For the electron microscopic examinations the samples were fixed in 1 per cent $KMnO_4$ and embedded in durcupan. Photosynthetic activity was demonstrated by $^{14}CO_2$ incorporation (spec. act. 0.4 mCi/nmol) at an illumination of 10 000 lux, after assimilation maintained for 1 hour. The chlorophyll content was determined by the method of ZSCHEILE—COMAR (1941). For detailed description of methods see SZUJKÓ-LACZA *et al.* (1971).

In order to gain information on age characteristics, one- and two-year- old shoot axes were collected, that is, axes, which had overwintered for a first or second time. Having counted the grana of the chloroplasts statistical analysis was performed on the basis of electron microscopic photos. The data were evaluated by a bifactorial analysis of variance (SVÁB 1967).

Results

In the course of electron- and light-microscopic studies made on the cortex of one- and two-year- old shoot axes pronounced structural differences were found between cells — even within the same cell-row in a given phase. As cells nearly the same in structure as under summer conditions could be found throughout the year, this heterogeneity gradually increased. In selecting the pictures and interpreting the material the aim was to show the structure occurring with the highest frequency at a given time. In many cases electron micrographs are supposed to reflect the reaction of the cell to the fixing solution in the given physiological state, rather than the actual structure. However, from the different responses given to identical fixation an indirect conclusion can be drawn on the original structure. In this the light-microscopic observation of living cells in hand sections was of great help.

Table 1

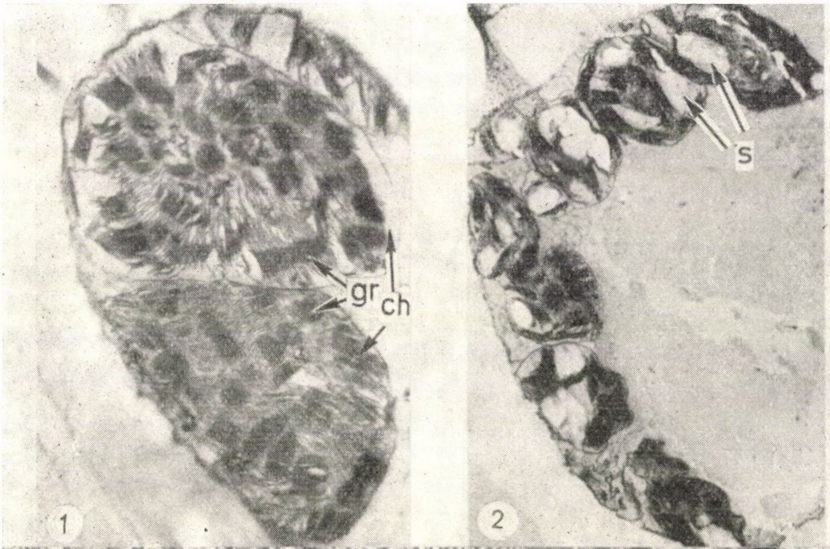
Seasonal changes in the photosynthetic activity of bark chloroplasts, as related to 10^{-4} mol CO_2/dm^2 bark surface

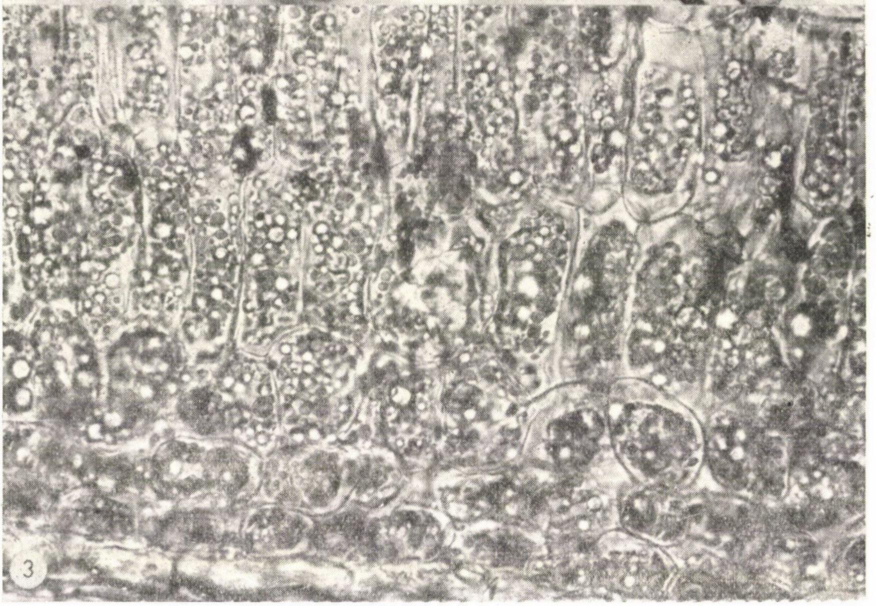
	July	October	February	April
Alcohol-soluble fraction	28	46	120	1
High molecular weight compound	6	21	284	14
Total activity	34	67	404	15

In July, chloroplasts in the chlorenchyma cells of the shoot axis are of granal structure, just like the leaves (Fig. 1) and may contain a large amount of starch (Fig. 2). The total amount of chlorophyll in the bark is only one-third of that in the leaf, but it is characteristic that the former has a relatively higher photosynthetic activity than the latter (Szujkó-Lacza *et al.* 1971). In summer the incorporation of $^{14}CO_2$ shifts toward the alcohol-soluble fraction (Table 1).

In October the vegetative period of *Euonymus europaeus* approaches an end, the leaves become yellow and the cells filled with oil (Fig. 3). From the outside the shoot axis is dark green, under a light-microscope the chloroplasts are slightly yellowish, the cuticle seems faintly thickened and the outlines of the plastids cannot be clearly seen in the chlorenchyma. In the proplastids of the phelloderm (Fig. 4), in various zones of the cortex, thus in the outer chlorenchyma cells as well (Fig. 5), and in the eight and ninth cell-rows large amounts of starch can be found. From the great number of mitochondria intensive oxidative processes can be concluded on. Protuberances can often be found on the side of the thickened cell-walls adjacent to the intercellulars (Fig. 6). A great many crystals accumulated in the whole of the bark (Fig. 7).

Within the total activity the alcohol-soluble fraction is still dominant, but the proportion of high molecular weight compounds has increased compared to that in July (Table 1). The total chlorophyll content shows an autumn minimum (Fig. 8).





Figs 1–2. (July) Chloroplasts with a great number of grana (Fig. 1, 13200 ×) and plastids containing much starch in the primary cortex (Fig. 2, 4400 ×)
Fig. 3. (October) Mesophyll-cells filled with oil in the leaf. 250 ×

cf: cytoplasmic fibrils; ch: chloroplast; cr: crystals; cw: cell-wall; ER: endoplasmic reticle; f: fusion of chloroplasts; gr: granum; l: lipid; m: mitochondrium; n: nucleus; pr: proplast; pt: protuberance; s: starch; v: vacuole

The January examinations aim at outlining the structural and certain functional conditions in the apparent dormancy period of the plant. Under a light microscope the cytoplasm in the cortical cells seems frozen and swollen and in most cases gets a uniform light green colour from the chloroplasts. The borders of the chloroplasts are generally difficult to distinguish. (These observations are described in detail by ALEKSANDROV—SAVCENKO 1950, and others.) There are, however, numerous cells in the tissue which have intact chloroplasts. According to the electron microscopic observations the epidermal cells maintain their internal structure indicating intensive functioning; the large amount of endoplasmic reticulum is especially remarkable in them, and they also contain much oil (Fig. 9). From the plastids of the primary cortex, especially from the outer cell-rows, the starch has totally disappeared. The central vacuole is divided into a number of smaller vacuoles (Fig. 10a),

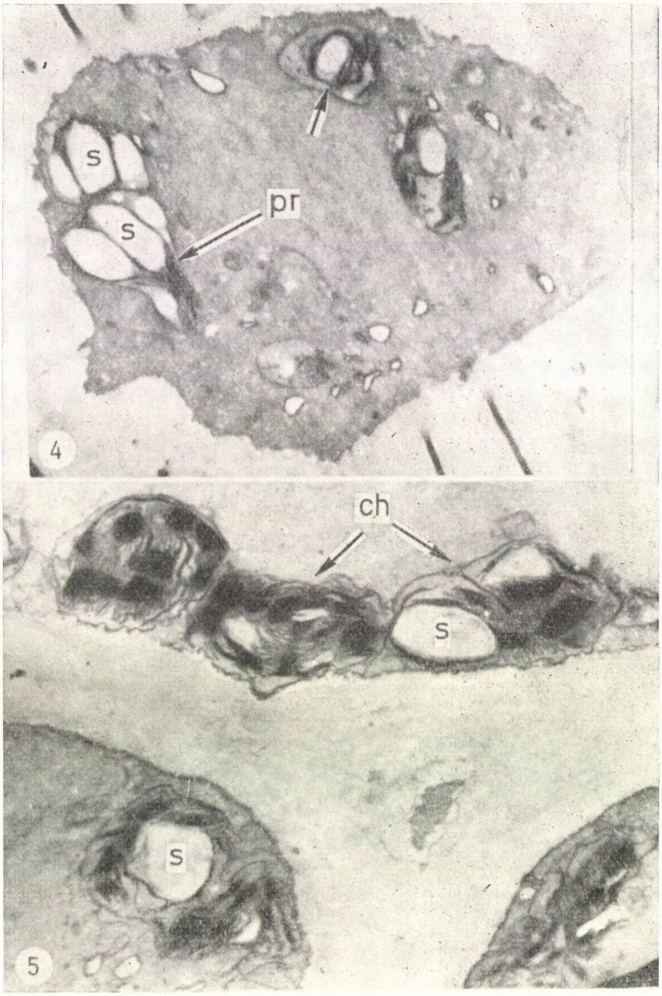
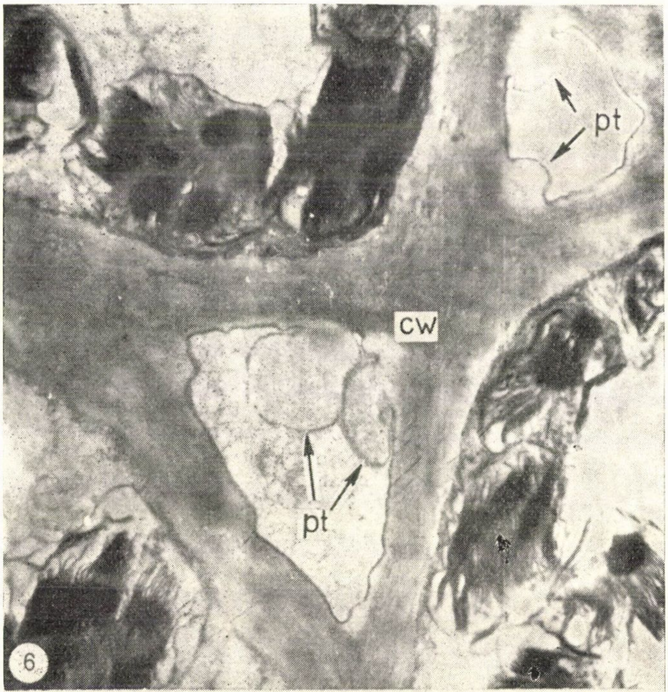


Fig. 4—5. (October) Proplasts in the phelloderm (Fig. 4. 4400 ×) and chloroplasts in the cortex filled with starch (Fig. 5. 8800 ×)



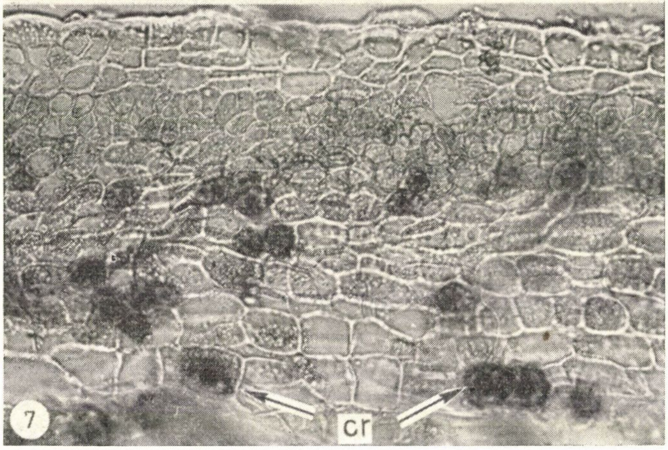


Fig. 6. (October) Cell-wall protuberances toward the intercellular space. 8800 ×
Fig. 7. (October) Crystal accumulation in the primary cortex. Tangential longitudinal section 100 ×

and consequently the otherwise swollen nucleus moves from the side of the cell-wall to the centre of the cell (Fig. 10b). The plastids are generally moved too, and grouped mostly around the nucleus (Fig. 10b), or by one side of the cell-wall. The lamellae of the chloroplasts become sparse; this can be observed even in those cells which otherwise maintain their original structure (Fig. 11).

Electron-dense granularity is often found in the chloroplasts of the two-year-old bark (Fig. 12). A considerable decrease is experienced in the number of grana. In January the cells contain an abundance of mitochondria.

At the end of February, beginning of March water circulation starts in the plant, the buds begin to swell. The nucleus is still in the centre in the chlorenchyma cells (Fig. 13), but the vacuoles begin to flow together, they decrease in number and increase in size until at last the thin threads of plasm break and the plasm lining along the cell-wall surrounds the restored large central vacuole. The chloroplasts move from the nucleus to the cell-wall and gradually develop into typical granal chloroplasts. Mitochondria occur in large numbers (Fig. 14).

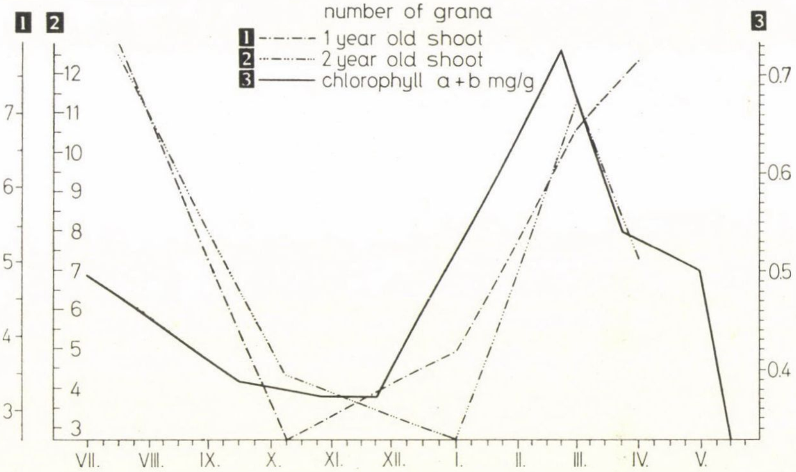


Fig. 8. (graph) Annual course of the number of grana and total chlorophyll content in the plastids of primary cortex cells

Photosynthetic activity — the trend of which has already been indicated by the October measurements — becomes reversed at the end of February compared to the summer conditions, and CO_2 incorporation shifts toward the high molecular weight compounds, that is, toward synthesis. Activity is higher than the summer value by one order of magnitude.* The annual maximum of total chlorophyll content is in this month. A most diversified picture is obtained with a light microscope at the end of February, beginning of March. The nuclei are mostly swollen, the chloroplasts are of various size, and the thick, aspic-like character of the cytoplasm begins to disappear. This transitional stage, which involves a conspicuous increase in activity, seems to be the most reactive phase of both cell and organism. This supposition is con-

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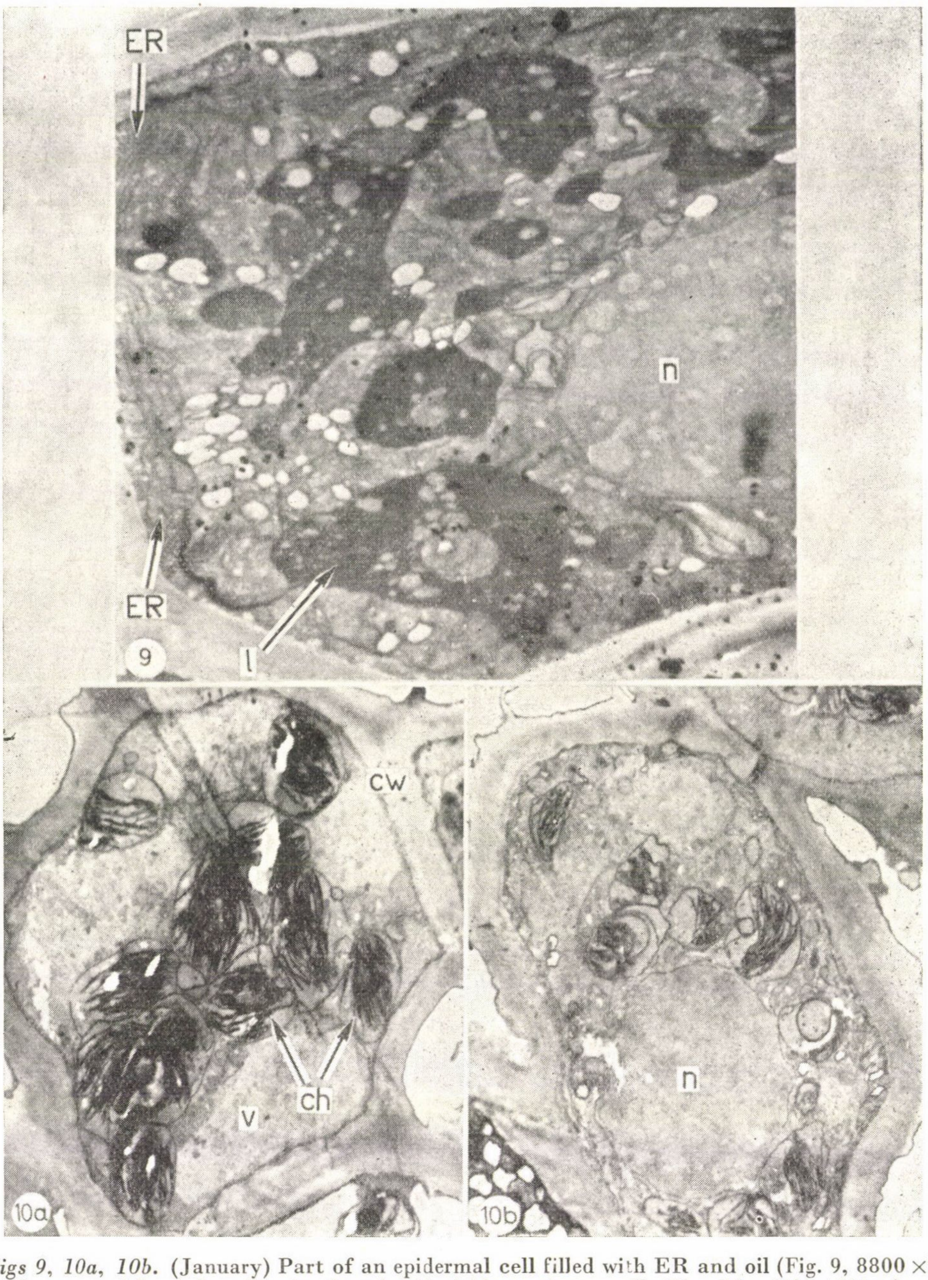
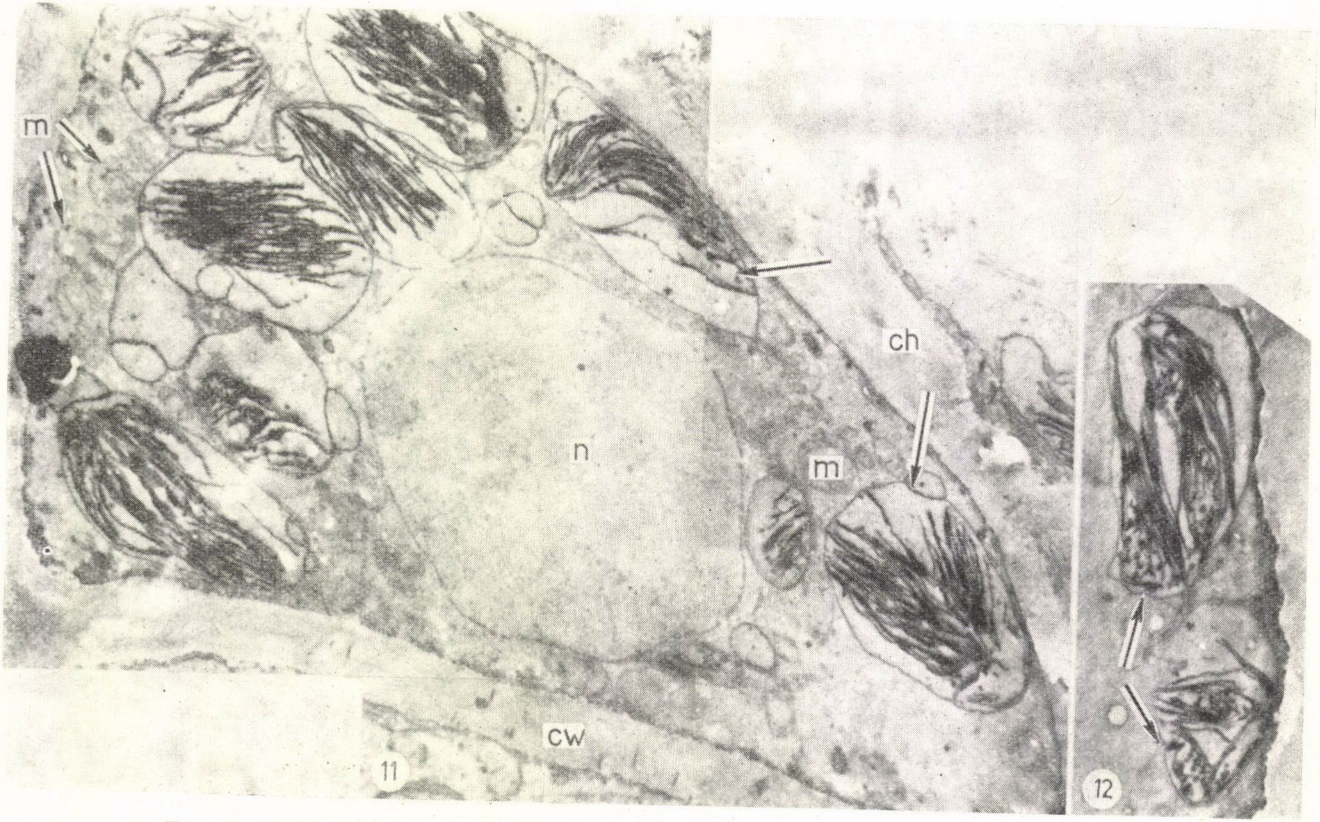
* The fact that the surprisingly high activity value in February is no error of measurement is confirmed by the similarly high photosynthetic activity of two other species: *Quercus pubescens* and *Fraxinus ornus* at that time.

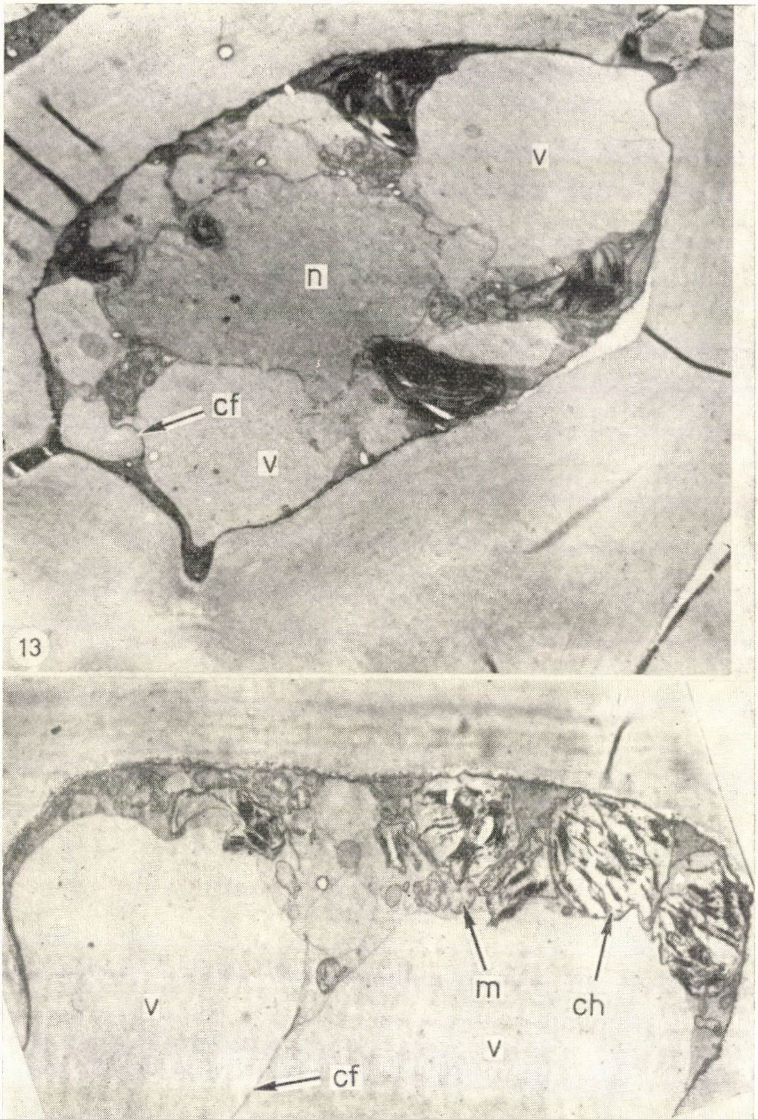


Figs 9, 10a, 10b. (January) Part of an epidermal cell filled with ER and oil (Fig. 9, 8800 ×). Sparse and loose lamellae in the plastids (chlorenchyma cells Fig. 10a, 10b, 4400 ×)

firmed by the fact that on the surface of hand sections prepared from shoots exposed to room temperature free green pigment can be seen supposedly originating from the injured chloroplasts and accumulating in the intercellular



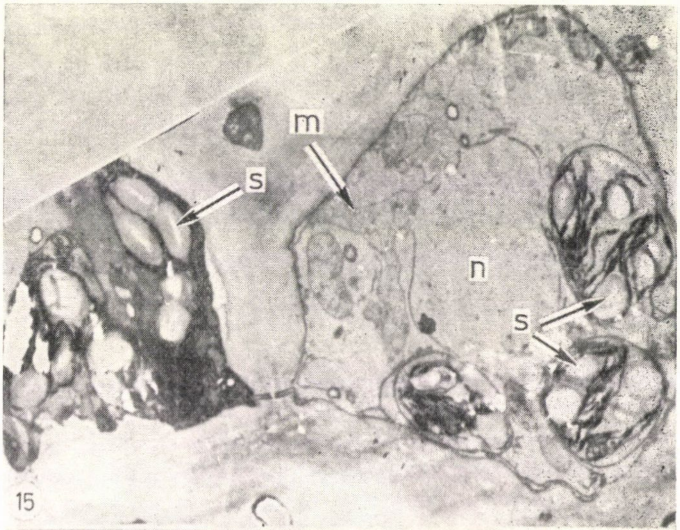
Figs 11, 12. (January) Slightly desorganized plastids, a lot of mitochondria in the otherwise unchanged chlorenchyma cell (Fig. 11) and electron-dense granularity in the plastids of the two-year-old shoot axis (Fig. 12, 8800×)

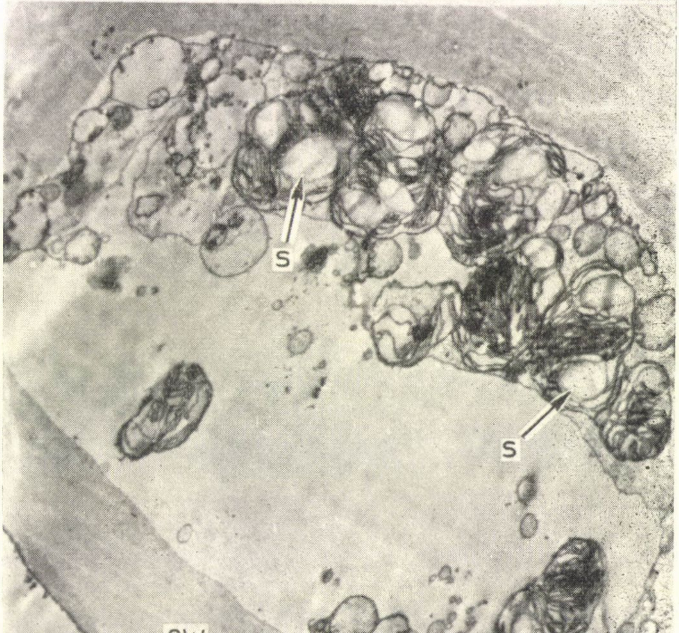


Figs 13, 14. (March) Plastids returned to the cell-wall (Fig. 13); central vacuole restored (Figs 13, 14, 4400 ×)

space. The intercellular space in the electron microscopic material fixed in this period is often filled with dense material.

April examinations were carried out when the young leaves were appearing. Simultaneously with the development of the leaves starch began to





Figs 15, 16. (April) Intensive starch accumulation in the phelloderm- and chlorenchyma cells (Fig. 15), and in the more inner cell-rows of the primary cortex (4400 ×)

accumulate in the phelloderm, in the outer and inner cell-rows of the primary cortex (Figs 15, 16). The number of grana was close to the July value. The chloroplasts gradually regained their original shape as confirmed unequivocally by the light-microscopic observations. In the course of the year total activity was the lowest in April, and its trend reflected the conditions of late February, while its proportion increasingly shifted towards the high molecular weight compounds. In April the total chlorophyll content showed a transition towards the minimum at the end of May.

It should be mentioned here, that during the electron microscopic examinations in January unusually large chloroplasts were observed which — owing to their size and structure — looked as if they had been made up of several smaller chloroplasts (Fig. 17). In April some chloroplasts desorganized which for the most part manifested itself in the swelling of the grana (Fig. 18). The two phenomena observed are supposed to be the result of some artificial product, however, this different reaction to the fixing solution may have a functional explanation as well.

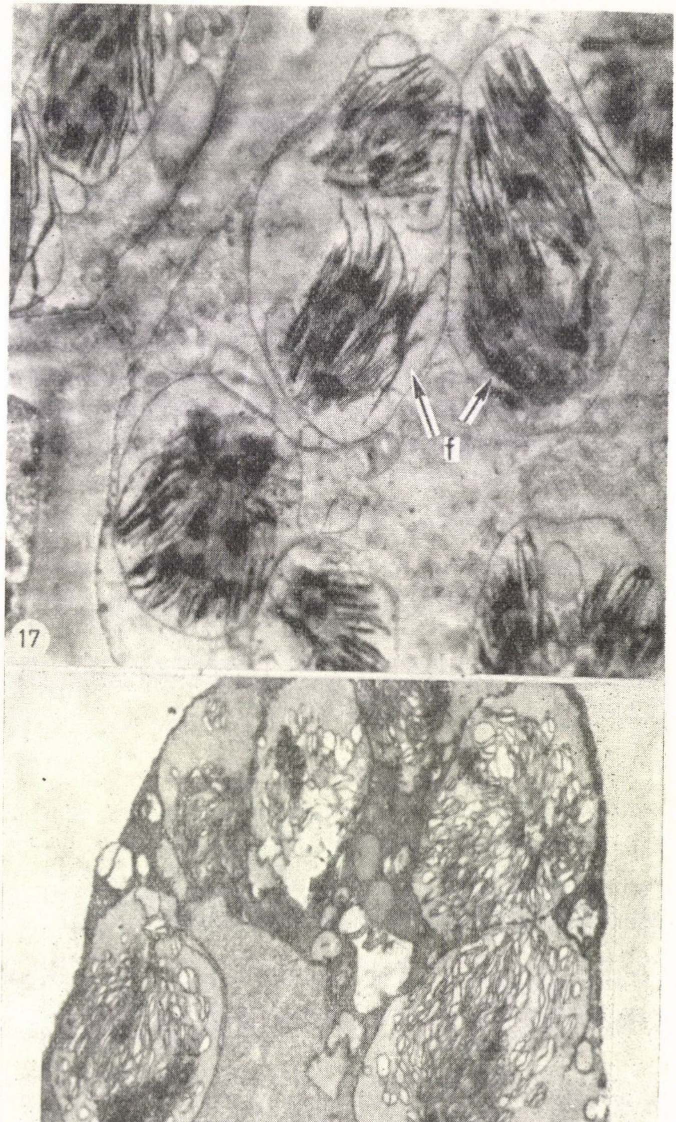
Table 2

Seasonal changes in the number of grana per chloroplast (each of the data is the average of 11 counts). S.d. 0.1% = 3.4

	July	October	January	April
One-year-old shoot axis	8.0	2.7	3.5	7.7
Two-year-old shoot axis	12.5	3.4	2.7	7.4

On the basis of preliminary studies it was found that among the structural elements of the chloroplasts it was the grana that were suitable for indicating numerically the seasonal changes in the chloroplasts. Therefore the effect of seasonal factors, and the age of the shoot on the chloroplasts were evaluated on the basis of changes in the number of grana. The authors' experimental setting (randomized design, bifactorial, two- and four-level analysis of variance) made it possible to assess the age of the shoot (one factor) and the seasonal abiotic effects (the other factor). (The levels are the two age-groups and the four months.) The result of the analysis is significant (P = 0,1%) and the effect of the two factors is respectively too. Both the chloroplasts, and the number of their grana in shoots of different age respond to the seasonal effects practically in the same way. With the same seasonal effect (in the same month) significant difference is found only once (July) according to the age of the shoots. By October the number of grana decreases to a minimum; remains at this level till January; in April it reaches or approaches the summer (July) level (Table 2).

Within the same month — if standard deviation is expressed as a percentage of the average (coefficients of variance) — one- and two-year old shoot axes do not show the same trends (March values are also considered here, Table 3).



Figs 17, 18. Melting of plastids in January, and desorganization of grana in April, 8800 ×

Table 3

Coefficient of variation of the number of grana (CV%)

Date	One-year-old shoot axis CV%	Two-year-old shoot axis CV%
1970 July	35.5	38.1
October	48.3	35.1
1971 January	42.9	28.8
March	20.6	33.0
April	34.8	32.9

Discussion

The cytoecological periodicity of *Euonymus europaeus* bark, green even in winter, was measured by the seasonal changes in the structure and function of the chloroplasts. Although in the vegetative period and so-called dormancy of the plant the chloroplasts maintain their size and fundamental inner structure, — in agreement with the results obtained by PARKER—PHILPOTT (1963)— their lamellar structure undergoes certain changes during the year. When studying chloroplasts in *Betula* and *Sambucus* barks KRASAVTZEV—TUTKEVICH (1971) found that in winter the lamellae became sparse. According to the authors' investigations too, the number of lamellae decreases in winter, but the seasonal effects can be best assessed by the changes in the number of grana (Fig. 18, Table 2).

The coefficient of variation values of the number of grana showed higher fluctuation in the one-year-old-shoot than in the chloroplasts of cortical cells in the two-years-old shoots (Table 3), which possibly means that both chloroplasts, and grana in the chlorenchyma cells of the two-years-old shoot have more uniform reactions.

The seasonal changes could be observed not only structurally, changes in the chlorophyll content reflected them well too. According to Fig. 8 the number of grana as well as the total chlorophyll content decrease unambiguously from July to October; starting from December—January the values in question gradually increase till March, the beginning of water circulation, and the total chlorophyll content shows the annual maximum. The number of grana reaches the July level. From March the total chlorophyll content gradually decreases till the annual minimum in May, while in July shows the summer- and annual second maximum. The parallel curves well reflect the connection between chlorophyll content and number of grana. The two observations carried out separately support each other. A similar change in the annual total chlorophyll content (winter maximum, second maximum in summer and early spring minimum) was observed by SCHENK (1952) in the bark which contains the primary cortex of the ecologically similar *Tilia cordata*. This similarity underlines the role of ecological factors in the annual trend of chlorophyll content.

The annual course of photosynthetic activity can be parallelled with the phenology of the plant (Table 1). The total activity of the chloroplasts in the bark suddenly increases from October till the end of February — till bud swelling — and exceeds the summer values by one order of magnitude, supposedly because the leaf as an organ producing organic matter is shed and certain photosynthetic processes are performed by the primary cortex. In early April the leaves begin to develop and display photosynthetic activity; then the activity of the bark suddenly decreases.

At the end of winter and early spring the synthesis of high molecular weight substances is dominant; they may play a role in producing energy for bud sprouting. When the leaves appear the functional importance of the bark decreases, so the function of the biosynthesis taking place here is to meet the requirements of the bark cells; at this time mainly the directly available low molecular weight substances are formed. Photosynthetic activity at the end of winter was proved again only in vitro, but the fact that the ability of incorporation could be realized at the growing site too was supported by the in vivo photosynthesis measurements carried out in winter, below freezing point (summing up v. STÅLFELT 1960). At the end of winter, in the open, during the melting period photosynthesis — a positive CO_2 balance — rapidly increases (TRANQUILLINI 1957, UNGERSON—SHREDIN 1965).

The authors followed the seasonal changes of starch in the primary cortex of one- and two-year-old shoot axes. They found that it was only from the outer zone and one or two cell-rows of the next zone that starch completely disappeared by January; in the more inner layers it remained intact or was mobilized later and only to some extent. According to PARKER (1958) and many others during the winter most of the starch is transformed into sugar, and the degree of sugar concentration influences the winter-hardiness. Winter-hardiness has extensive literature in which it is explained partly by the increase in the sugar concentration, partly by the accumulation of protein and RNA and swelling of the nucleus, partly by the sol-gel condition of the cytoplasm (BIEBL 1962, PARKER 1958, SIMINOVITCH 1963, KRASAVTZEV—TUTKEVICH 1971, TUMANOV 1967, LYR—POLSTER—FIEDLER 1967).

Cytological adaptation to changed environmental conditions was only studied on a morphological level. In the course of electron microscopic observations it was found that by winter the large central vacuole became divided. In summer, on the other hand, the original state was restored. Changes of cytoplasm in winter are attributed partly to plasmolysis (pseudo-plasmolysis, BIEBL 1962), partly to vacuolization. The first case may be caused by ice formed in the intercellular space, the latter phenomenon is interpreted by two hypotheses: a) vacuolization occurs in cells in the case of starch deficiency, b) the phenomenon is considered as some sort of cell regeneration, re-meristematization (Giese 1957, Gill—Vear 1958).

According to the authors' investigations the chloroplasts of *Euonymus europaeus* primary cortex cells maintain their shape, structure and chlorophyll content in a more or less stable form throughout the whole year, and are able to display photosynthetic activity all the year round or even for several years. Inasmuch as their presence is not considered to be atavism, they are supposed to take part in the life of the plant in a way and extent outlined above.

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NUCLEAR DEPENDENCE OF HORMONE RESYNTHESIS IN THE ADRENOMEDULLARY CELLS OF RATS

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Rats were treated with 5-fluorouracil or actinomycin D, in order to examine the function of altered nucleic acid metabolism in the process of catecholamine resynthesis following insulin-induced catecholamine secretion. Insulin (20 IE/100 g body weight) administered i.p. caused marked catecholamine secretion. 24 hours after insulin treatment, the catecholamine content of the adrenal gland was only half of that in the untreated control. 168 hours after the insulin was given, the catecholamine level had returned to normal. If 5-fluorouracil or actinomycin D was injected 6 hours after the insulin, resynthesis of catecholamines was strongly inhibited. Thus, after 168 hours the adrenomedullary catecholamine content in doubly-treated animals was only half that of insulin-treated controls. As hormone synthesis takes place in the cytoplasm of the cells and the primary effect of 5-fluorouracil and actinomycin D is on the cell nucleus, it would seem that the process of cytoplasmic hormone synthesis can take place undisturbed only if nuclear metabolism is intact, i.e. the process is nucleus-dependent.

The "synthetic" and "anabolic" role of the nucleus in cell metabolism was suggested rather a long time ago (Mazia 1952). One or two decades ago however, the literature dealing with the mechanism of secretion (Palay 1958, Palade—Siekevitz—Caro 1962) only referred occasionally to the role of the nucleus. In those day investigations mostly aimed at elucidating the importance of the cytoplasm and of cytoplasmic cell organelles in the process of secretion. Although one finds references to morphological changes in the nuclei encountered in the course of studies on cytological alterations taking place during secretion, the explanations offered concerning the physiological importance of these alterations are only tentative (Kurosomi 1961).

In our electron microscopic studies on induced catecholamine secretion of the adrenal medulla (Benedeczky 1968) we have observed various nuclear alterations. These alterations suggested that in the adrenal medulla there might be a direct relationship between the secretory activity of the gland and the metabolism of the nucleus. Besides morphological observations carried out by means of electron microscopy, the results of cytological examinations also speak in favour of this theory. Thus in chromaffin cells, cell division was not observed during resting secretion (Viola-Magni 1966) an observation which also supports the suggestion that the nucleus might affect the process of secretion principally by influencing cell metabolism.

These considerations led to experiments in which rats were treated with 5-fluorouracil or actinomycin D, in order to examine the function of the altered nucleic acid metabolism in the process of catecholamine resynthesis, following insulin induced catecholamine secretion. In this way it was hoped to obtain direct evidence for the role of the nucleus in secretion. Actinomycin D and 5-fluorouracil were chosen because the primary effect of these drugs is on the cell nucleus.

Material and Methods

120 laboratory Wistar rats of both sexes (average weight 200 g) were used. Prior to treatment the animals were deprived of food for 24 hours, but drinking water was available ad libitum. Insulin was administered intraperitoneally at a dose of 20 IE/100 g body weight. No signs of shock condition were observed following this treatment. Six hours after injecting the insulin, the animals were given 5-fluorouracil at a dose of 250 mg/kg or Actinomycin D at a dose of 250 μg/kg. This low dose of Actinomycin D was chosen because it did not cause any obvious stress on the animals. Only insulin-treated animals were used in examinations of the chronological process of catecholamine (C.A.) resynthesis. The animals were killed by decapitation at various intervals — at 6, 24, 48, 72, 96, 120, 144 and 168 hours, following injection of insulin, and the adrenal glands were immediately removed and weighed. The amount of total catecholamines and adrenaline was determined by the Euler—Hamberg method as modified by Puppi *et al.* (1959). From the measured values we have calculated the amount of noradrenaline (noradrenaline = total catecholamine—adrenaline) and the ratio of adrenaline to noradrenaline. Photometric measurements were performed using a Unicam spectrophotometer at a wavelength of 529 nm.

Results

In the experimental group treated with only insulin, as early as 6 hours following treatment the total catecholamine level had fallen to an average value of 800 μg/g, from the value of 1400—1600 μg/g seen in untreated animals (Fig. 1). Catecholamine content was lowest 24 hours after treatment, when the total catecholamine content of the adrenal medulla had dropped to 680 μg/g. After 48 hours, as a sign of the commencement of resynthesis, the catecholamine content increased to 890 μg/g, rising to a value as high as 1170 μg/g after 72 hours. From this time on, until the end of the experimental period, the total catecholamine level increased slightly and ranged within that of the control group not treated with insulin. The ratio of adrenaline to noradrenaline (A : NA) was; 63 : 37 at 6 hr, 58: 42 at 24 hr, 64; 36 at 48 hr, 72; 28 at 72 hr and remained essentially unchanged in the later period of the resynthesis.

In the experimental group treated with insulin plus actinomycin D the total catecholamine content decreased to 800—900 μg/g in the period 6—24—48 hours following the administration of the drugs (Fig. 2). From 72 hours to 168 hours the total catecholamine content of the adrenal medulla decreased further to about one-third of the control (at 96 hours 460 μg/g.)

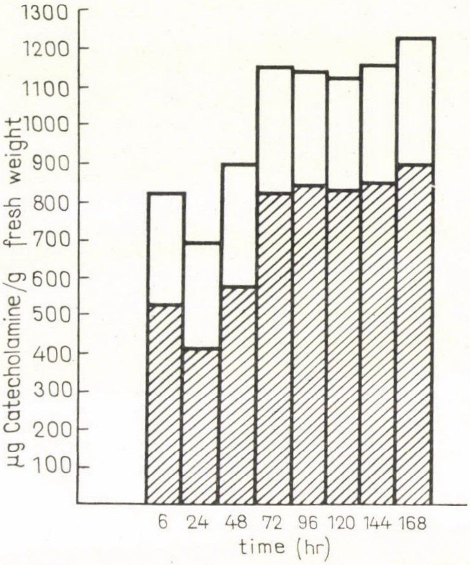

Fig. 1. The change of catecholamine content of the adrenal gland following insulin treatment. The columns show the total catecholamine content in which the adrenaline content is represented by the shaded areas and the noradrenaline content is indicated by the free areas

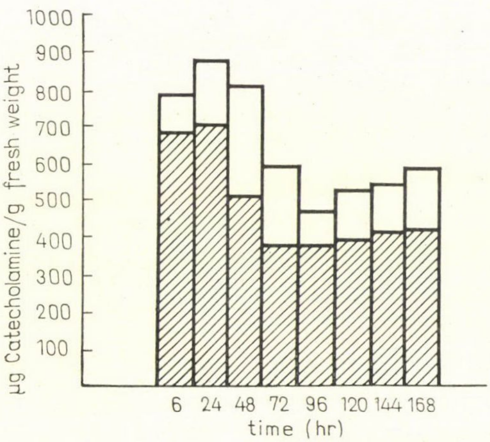

Fig. 2. The change of catecholamine content of the adrenal gland following insulin + actinomycin D treatment

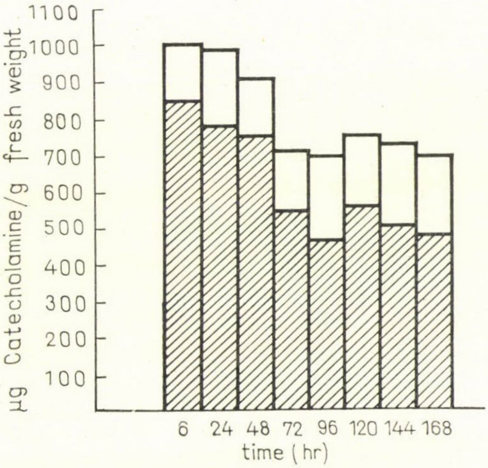

Fig. 3. The change of catecholamine content of the adrenal gland following insulin + 5-fluorouracil treatment

In rats treated with insulin and actinomycin D, the ratio A: NA was 62: 38 after 48 hours and 64: 36 after 72 hours.

In the group treated with insulin plus 5-fluoruracil, total catecholamine content showed similar changes to those observed following combined treatment with insulin and actinomycin D (Fig. 3). From 72 to 168 hours, however, the inhibition of resynthesis appeared to be somewhat less pronounced and the amount of catecholamine was about 700 $\mu g/g$ in this period. In contrast to observations made during actinomycin D treatment, the ratio A: NA did not shift in favour of NA, following treatment with 5-fluorouracil (at 48 hr = 83 : 17, at 72 hr = 78 : 22).

Discussion

The few studies dealing with the role of the nuclei of adrenal medullary cells in the process of secretion have yielded contradictory results. According to LEEMAN (1959, 1959a) exposure to cold does not influence the volume of the nucleus; however it does increase the DNA content. On the other hand, a decrease of about 20—40 per cent in DNA content was observed by VIOLA-MAGNI (1966) also after cold stress. ROELS (1963) registered a marked increase in the dry-matter content of the nuclei as a result of chronic cold stress, while shrinkage of the nuclei subsequent to intensive secretion, and an increase in volume of the nucleus during long-lasting (but not intensive) secretion was reported by SÉTÁLÓ (1962).

It can be stated, on the basis of the observations just described that the nucleus may undergo various changes dependent on the secretory state of the gland. However, the nature of these alterations is not clear, and the alterations produced do not provide sufficient information of either the nature of the nucleo-cytoplasmic interaction or the probable role of cell nuclei in secretion.

In our examinations we have set out to create an experimental model which might suitably be used to investigate these problems. The applied insulin load provoked marked catecholamine secretion and hormone resynthesis. The process of hormone resynthesis was similar to that described by HÖKFELT (1951). It was expected, on the basis of their well-known cytostatic effect (HEIDELBERGER 1963, PERRY 1963, LAPIS—BENEDECZKY 1966, BERNHARD—GRANBOULAN 1968, STENRAM 1969, BENEDECZKY—KOPPER 1969) that 5-fluorouracil and actinomycin D might produce specific changes in the nucleic acid metabolism. Indeed, we noted morphological signs of this (formation of "spotted" nucleoli, nucleolar segregation, karyolysis and pyknosis, increased transparency of the karyoplasm, appearance of quadrilamellar membranes, etc.) in an earlier electron microscope study (BENEDECZKY 1968). In the course of our ultrastructural electron-histochemical and autoradiographic studies (BENEDECZKY—KOPPER 1969) of "spotted" nucleoli, we arrived at the conclusion that parallel to the appearance of "spotted" nucleoli, following 5-fluorouracil treatment, the RNA synthesis of the cells decreased and, simultaneously, the rate of nucleolar labelling increased. In the present study we did not wish to engage in a detailed analysis of the changes in nucleic acid metabolism; we wanted only to register some of the alterations produced, and the quantitative measurements were concerned with the catecholamines. The data obtained show that if 5-fluorouracil or actinomycin D was applied in the early phase of hormone resyntheses, the synthetic process was permanently and significantly inhibited. Because hormone synthesis takes place in the cytoplasm of the cells, and the primary effect of these drugs is on the cell nucleus, it would seem that the process of cytoplasmic hormone synthesis can take place undisturbed only if nuclear metabolism is intact, i.e. the process is nucleus-dependent.

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VEGETATIVE CELL NUCLEUS, GENERATIVE CELL OR MICROGAMETES ENTERING THE POLLEN TUBE IN CONSOLIDA AJACIS (L.) SCHUR.

By

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The nucleus of the vegetative cell is not always the first to enter the pollen tube of *Consolida ajacis* (L.) Schur. when germination begins. The generative cell or the two microgametes, respectively, may also enter first. In microgametes entering the pollen tube and advancing in it no active motion can be demonstrated. With the growth of the pollen tube progressing the pollen grain becomes empty, and the nucleus of the vegetative cell as well as the microgametes get into the pollen tube in each case. In the pollen grain and pollen tube the microgametes change place — in our opinion — by the cytoplasm flow of the vegetative cell of the microspore, or through the orientated and systematic contraction of cytoplasm tubules included in the cytoplasm.

Introduction

Cells and cell organelles contained in the microspore get into the growing pollen tube in the course of tube development. Among the cell organelles the nucleus of the vegetative cell was thought to be the first to enter the pollen tube, since regulating and orienting roles were attributed to it in the process of tube development. After the two- and three-nucleate microscopes had been discovered — which were considered as characteristic of the species — it was thought that the nucleus of the vegetative cell got into the growing pollen tube first only in the case of the two-nucleate pollen grain; while in the case of the three-nucleate microspore it was the nuclei of the generative cell that first entered the pollen tube, and the nucleus of the vegetative cell was for some time retained in the pollen grain — until it was emptied. On the other hand, POLYAKOVA (1958), POLYAKOVA—PODSTAVEK (1960), RUDENKO (1956) found that in the two-nucleate *Tradescantia fusca* L. and three nucleate *Hesperis matronalis* L. the nucleus of the vegetative cell got into the pollen tube in every case irrespective of whether the microspore was two-nucleate or three-nucleate. KIHARA—HORI (1966) pointed out that it depended on the species which of the nuclei entered the pollen tube first, as in wheat and rice the nucleus of the vegetative cell was often found to remain in the pollen grain, while in maize such a case never occurred. Later it was discovered that the cytoplasm of the vegetative cell was separated by a double unit membrane from the two generative nuclei in the pollen grain and in the pollen tube too (HASSENKAMP 1960). The existence of this separating layer was proved by experiments carried out with different osmotic pressures used (HOFFMEISTER 1956), and it was found to become similar to the meristemic cell wall during the process of pollen maturation (SASSEN 1964). Thus, the generative cell nuclei — by possessing independent and separate cytoplasms — are equivalent to other cells. So the supposed importance of the vegetative cell nucleus in controlling the growth of the pollen tube decreased. In the case of *Tulipa gesneriana* L. HERICH (1969) pointed out that the nucleus of the vegetative cell is not always the first to enter the pollen tube; the generative cell and the microgametes may get there first too. He says further, that it is not immediately at the beginning of tube formation that the nucleus of the vegetative cell enters the developing pollen tube. And even if it gets there first, it may not occupy the tip of the pollen tube but is localized considerably farther inside. From these observations the author drew the conclusion that the nucleus of the vegetative cell did not take part directly and actively in controlling the growth of the pollen tube.

The nucleus of the vegetative cell, the generative cell and the microgametes have no motional organs, still they get from the pollen grain into the pollen tube. The nucleus of the vegetative cell, as a cell organelle can get into the pollen tube only in a passive way, through the flow of the cytoplasm (cyclosis) or contraction of the cytoplasm tubules. Development of pseudopodia, though also known in the nuclei of cells only occurs in cell cultures and in fungi. As for the generative cell and the microgametes getting into the pollen tube, two opinions exist. According to the opinion of KOSTRYUKOVA—BENETSKAYA (1958) — which agrees with NAVASIN's hypothesis — the generative cell, and the microgametes respectively, get into the pollen tube by moving actively by themselves; while KIHARA—HORI (1966) suggest that these cells are carried passively, by the flow of the cytoplasm into the pollen tube.

Material and Methods

Investigations were made to find out in an indirect way whether the generative cell, and the microgametes — which are equivalent to other cells — respectively, get into the pollen tube in an active or passive way.

The pollen grains of *Consolida ajacis* (L.) Schur. were germinated on a culture medium prepared from a mixture of 5 cm^3 16 per cent sugar solution and 5 cm^3 A-Z solution. The optimum sugar concentration was determined from the result of pollen germination carried out on culture media containing various amounts of sugar; in the case of the given plant species it was an 8 per cent sugar solution. Tube formation of pollen grains also occurred in sugar solutions not containing A-Z solution but it was considerably slower and of a lower extent than that observed on a culture medium containing trace elements.

A drop of the culture medium was spread over a slide and sprinkled evenly with pollen from a mature anther. These slides were put into Petri dishes in which cotton wool saturated with hot water had been placed previously. Thereby the pollen grains were placed at once at a constant temperature and in an atmosphere saturated with humidity.

Pollen grains that had developed tubes were stained with carmine acetic acid, because in studies made on the process of microgametogenesis this method of staining proved better than the Feulgen technique. With carmine acetic acid used for staining under the influence of heating the cytoplasm of the cell becomes pink and the nuclei take on a bright red colour. Microscopic examinations were carried out by means of a Leitz Ortholux microscope on several thousand slides.

Direct investigations were made to find out whether it was the nucleus of the vegetative cell or the generative cell, and the microgametes respectively, that were the first to enter the pollen tube most often at the beginning of pollen germination.

In our studies we had the following idea: 1) If there is no plasm flowing into the pollen tube and if the microgametes move actively, then the latter get into the pollen tube before the nucleus of the vegetative cell which has no ability of active motion. 2) If there is plasm flowing into the pollen tube, and the nucleus of the vegetative cell is carried in by the current of the plasm, even then it is the microgametes that first get into the pollen tube provided that they move actively. Namely, the direction of the plasm current agrees with the way by which the microgametes get into the pollen tube, so their speeds should sum up, and again it is the microgametes that first enter the pollen tube.

From the frequency at which the nucleus of the vegetative cell and the microgametes first enter the pollen tube conclusions can be drawn concerning the active motion of the microgametes. Namely, if the nucleus of the vegetative cell is the first to get into the pollen tube, then the active motion of the microgametes is improbable; if, on the other hand, it is the microgametes that in most cases enter the pollen tube first, then this phenomenon makes their active motion probable.

Results

The pollen grains of *Consolida ajacis* (L.) Schur. are filled up mostly by the vegetative cell and its cytoplasm respectively. The cytoplasm of the generative cell is small and both in the mature pollen grain and in the pollen tube is located in they cytoplasm of the vegetative cell. It is in every case the cytoplasm of the vegetative cell that flows into the growing pollen tube; it becomes pink when stained with carmine acetic acid. After the cytoplasm has got into the tube it is either the nucleus of the vegetative cell or the generative cell — and the two cell-equivalent microgametes respectively — that first enter the pollen tube. Namely, in the case of *Consolida ajacis* gamete formation i.e. the second division of the microspore may take place either in the pollen grain or in the pollen tube, that is, the place of the second division is not determined.

After the cytoplasm of the vegetative cell has got into the pollen tube it is in most cases the nucleus of the vegetative cell that first enters the pollen tube. When the nucleus of the vegetative cell precedes the generative cell, and the microgametes respectively, in entering the pollen tube, it occupies the frontal part though not always the tip of the pollen tube. As the pollen tube grows it moves together with the advancing cytoplasm. In this case the staining of the nucleus of the vegetative cell changes during the growth of the pollen tube. The nucleus of the vegetative cell was more intensively stained before the division of the generative cell than after. This decrease in the intensity of colour can only be observed when the division of the generative cell, and formation of gametes respectively, takes place in the pollen tube.

It does happen — though less frequently — that the generative cell is the first to enter the pollen tube. In this case during the growth of the pollen tube the generative cell, then the two microgametes produced occupy the frontal part of the pollen tube, and maintain this position throughout the growth of the pollen tube; the nucleus of the vegetative cell gets into the pollen tube after the generative cell. The nucleus of the vegetative cell displays a decrease in the intensity of colour, in this case too, after the formation of the gametes, though — according to our observations — this decrease is not of the same extent as in the case when the nucleus of the vegetative cell occupies the frontal part of the pollen tube.

It occurs — though not often — that — if the second division of the microspore i.e. the process of gamete formation has already taken place in the pollen grain — the two gametes get into the developing pollen tube first. In this case they too occupy the frontal part of the pollen tube and remain constantly there throughout the growth of the tube. In the vegetative cell hardly any decrease in the colour intensity of the nucleus can be observed.

If the generative cell is the first to enter the pollen tube it may happen — though very seldom — that the generative cell and the nucleus of the vegetative cell are located side by side in the pollen tube parallel with its longitudinal axis. In the case of a large number of examinations specimens can be found where the nucleus of the vegetative cell is located immediately in front of or behind the generative cell. The nucleus of the vegetative cell generally shows a lower colour intensity when located in front of the generative cell than when located behind it.

No such case was found in which the nucleus of the vegetative cell remained in the pollen grain. Namely, during the growth of the pollen tube the pollen grain is completely emptied, no cytoplasm remains in it. Even the pollen tube will not be completely filled with cytoplasm, the latter occupies a certain place behind the tip of the tube. When the cytoplasm advances in the newly developed pollen tube, walls forming a right angle to the length of the tube develop and close in the advancing cytoplasm from behind.

Conclusions

Our investigations support HERICH's (1969) results namely, that the nucleus of the vegetative cell is not necessarily the first to enter the growing pollen tube. On the basis of our observations we share KIHARA—HORI's opinion (1966) that neither the nucleus of the vegetative cell nor the generative cell are capable of active motion when getting into the pollen tube and advancing in it. They are carried into the pollen tube by the flow of the plasm, and generally the one that has first entered leads the way within.

It was supposed that the microgametes move actively by themselves. Such a motion may be either an amoeboid- or a ciliary motion. Amoeboid motion is not probable in this case because its precondition is the possibility of adhering to a solid surface, which is unimaginable in the pollen grain and pollen tube, unless the microgamete passes along the wall of the pollen tube which is not probable either. Pseudopodia may be produced by both the nucleus and the cells even in liquid media. It is possible that the requirement of a solid surface is only relative, and the oriented change of viscosity occurring in the cytoplasm provides a solid surface for the amoeboid motion. However, if this process occurs at a right angle to the direction the microgamete moves in the motion mechanism of the cytoplasm too the change of viscosity, the reversible sol–gel transformation is one of the most important factors. Ciliary and flagellary motions — while adapted to liquid media — are excluded in this case, because microgametes have no motional organs. The cytoplasm motion of the microgametes does not result in any change of place, still during the process of fertilization they have to leave the pollen grain and reach the egg-cell and the vegetative cell of the embryo sac. In our opinion in the pollen grain and pollen tube the microgametes change place either by the cytoplasm flow of the vegetative cell of the microspore, or by the oriented and systematic contradiction of the cytoplasm tubules contained in the cytoplasm.

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DRAINAGE NEEDS OF RICE

By

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In green house studies on drainage, it was observed that surface as well as vertical drainage treatments were at par but were both superior to no drainage treatment in their effect on grain and straw yields. Better growth of plants in both drainage treatments could be attributed to a greater number of productive tillers and larger ear heads.

Introduction

It is a controversial point whether in the practice of land submergence maintenance of still water or moving water is more effective. The water can be kept moving by horizontal drainage — what is populary known as surface drainage — or by keeping it flowing. Alternatively, water can be kept moving by vertical drainage, viz. by allowing deep percolation. The function of both horizontal and vertical drainage is the same.

The available works on the above themes are summarized below. Some workers drained their fields during land submergence practice and claimed beneficial effects (**Baba** 1951, **Morrison** 1953, **Hayashi** *et al.* 1960, **Kawata—Ishihara** 1961, **Yamada** *et al.* 1961). In the opinion of other workers drainage reduces the yield or has no advantage (**Aglibut—Hoff** 1956, **Oyama** 1958, **Ueda—Oyama** 1958, **Nojima—Tanaka** 1961, **Tanaka** *et al.* 1961, **Aglibut** *et al.* 1962, **Nojima** *et al.* 1962).

It appears that three factors play a part in drainage effects. These are the soil type, the crop stage at which drainage is carried out, and the atmospheric temperature.

This trial was conducted with the object of finding out whether surface drainage had any advantage over vertical drainage, or whether maintaining continuous land submergence could result in obtaining maximum yield.

Material and Method

Earthen pots of 30 × 30 cms in size were used in this experiment.

Treatments: A — Continuous submergence.
B — Surface drainage.
C — Vertical drainage.

Design: Completely randomized with 13 replications. Small holes were bored in treatment C and these were fitted with corks to effect vertical drainage whenever necessary.

Each pot was filled with 7 kg of soil after mixing with it 1.01 g of ammonium sulphate, 0.83 g of single superphosphate and 0.28 g of muriate of potash to supply 60 kg N, 40 kg K_2O/ha. The quantity of water necessary to bring the soil to puddling was added and the pots were kept for setting. A 40 days old N.P.130 rice seedling was transplanted in each pot. Free standing water was 5 cm above the surface of the soil in the pots.

In treatment A during the growing period the measured quantity of water was added every day to maintain the submergence of 5 cm standing water. In treatment B, water was drained off from the surface by using rubber tubes and fresh water was added up to 5 cm standing water above the soil surface. In treatment C, standing water was allowed to drain by removing the cork at the base of the pot and fresh water was added to restore the same level as in treatments A and B. This was carried out daily from the date of transplanting till the maturity of the crop.

Results

The results of periodic observations on height and tiller numbers are depicted in Fig. 1. The plant height was more or less equal in the different drainage treatments. However, in the drained treatments a higher number of tillers were recorded than in the no drainage treatment.

Table 1 shows that the yields of grain and straw were the lowest in treatment A receiving no drainage at all. The lowest yield in treatment A could be attributed to the least number of productive tillers. The differences between other characteristics were not significant.

Table 1

Effects of submergence with surface/vertical and no drainage on plant characteristics

Plant characteristics	Treatments			SEm	C.D. at 5%	Results
	A	B	C			
Plant height (final in cm)	90.4	93.3	94.0	±0.38	1.1	*C B* A
Tiller number	12.5	14.6	14.9	±0.19	0.55	*C B* A
Productive tillers	12.3	14.4	14.6	±0.21	0.60	*C B* A
Ear head length (cm)	20.4	29.6	29.5	±0.17	0.50	*B C* A
Number of filled grains/ear head	179.4	179.8	179.3	±0.90	—	—
Unfilled grains/ear head	11.1	11.0	11.1	±0.28	—	—
1000 grain wt (gm)	18.8	18.9	18.7	±0.02	0.05	B A C
Grain yield/pot (gm)	31.4	34.4	33.2	±0.50	1.40	*B C* A
Straw yield/pot (gm)	51.9	54.2	53.9	±0.58	1.70	*B C* A
Protein % in the paddy grain	8.47	8.40	8.10	±0.09	0.27	*A B* C

A — Continuous submergence
B — Surface drainage
C — Vertical drainage

The treatments receiving vertical drainage and surface drainage were at par in respect to their effects on all plant characteristics except the 1000 grain weight. The 1000 grain weight was greater in treatment B receiving surface drainage than in that receiving vertical drainage. The grain and straw yields in the different treatments are shown in Fig. 1.

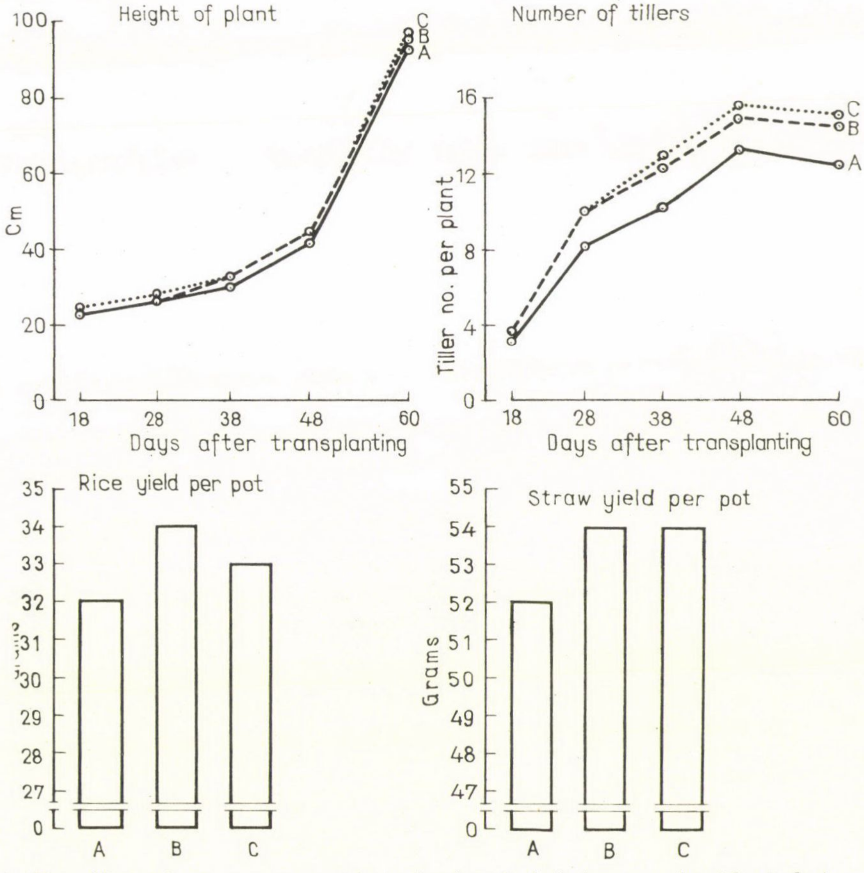

Fig. 1. The effect of submergence with surface/vertical drainage and without drainage on height of plant, number of tillers, grain and straw yield of N.P.130 rice. (A: — — — Submergence with no drainage. B: —— Submergence with surface drainage. C: . . . Submergence with vertical drainage.)

Discussion

The above studies on the effect of surface vertical drainage and no drainage have revealed an interesting observation about this practice.

The yields in treatments B and C were higher than in the no drainage treatment (A) on account of the higher values of all the yield attributing characteristics. The benefits of drainage are attributed to better aeration and/or removal of toxic substances (Fe^{++}, H_2S etc.) (KANNO 1956, VERMAAT 1950).

In these investigations the pots in the B and C treatments were drained daily. This would have resulted in maximum loss of nutrients possible under these situations. In spite of this loss, these treatments yielded more than treatments A having no drainage. These pots were 30 cm deep. Since rice has a well developed aeration system in its vascular bundles, it is surmised that adverse effect in treatment A resulting in lower yield may be due to the accumulation of toxic substances rather than lack of aeration. Further work, however, is needed to confirm this view.

Whether surface or vertical drainage is better will depend upon a given situation and normally there may not be any choice left in this respect. If there is a hard pan or rock present and the percolation rate is lower than 2.5 mms/day, it would be worth while to practice surface drainage. On the other hand, when the percolation rate is greater than 2.5 mm/day, natural percolation would be adequate to reap the advantages and there may not be any need for practising surface drainage.

In acid sulphate soils, vertical drainage may result in the formation of sulphuric acid through oxidation of iron sulphates in the soil. Similarly, sensitivity to drainage has been reported in peaty soils (Kari) in Kerala (SUBRAMANIAM 1952). Under such conditions, surface drainage would be preferable to vertical drainage (MOORMANN 1963).

Protein percentage was significantly higher in A and B treatments than in C having vertical drainage. The low protein percentage in C is probably due to leaching loss of nutrients. This result supports the view of NOJIMA *et al.* (1962) who have reported a decrease in the nutrient uptake of the rice plant at later stages of growth, because long time drainage decreases the availability of N, P_2O_5, SiO_2 etc.

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METAXENIE STUDIES OF PEAR VARIETIES

By

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Pollination studies were conducted between 1967 and 1970 with a total of 645 combinations, each year with a different number of varieties. According to the pomometric measurements made on ripe fruits, statistically proved metaxenic changes in the shape and size of fruits appeared in all three years in the following combinations: Vilmos (♀) × Bosc (♂), Vilmos (♀) × Pringalle (♂), Vilmos (♀) × Dupuit (♂); Hardenpont (♀) × Pringalle (♂), Hardenpont (♀) × Vilmos (♂), Hardenpont (♀) × Clapp (♂). Fruit colour metaxenie was obtained with the combinations Hardy (♀) × Vilmos (♂) and Hardenpont (♀) × Clapp (♂). As to the time of ripening, fruits of the combination Hardenpont (♀) × Pringalle (♂) attained picking ripeness 5–7 days earlier than the hybrid fruits of all the other combinations.

Introduction

Metaxenie is a result of breeding when the influence of alien pollens also shows in the fruit formation belonging to the mother plant. Metaxenie may appear in morphological forms (size, shape, colour, etc.) and in the form of physiological changes (in components, ripening time, storability, etc.), which develop a paternal character.

Metaxenic changes observed in pears are reported by HORN (1927), KIM (1946), NAGY (1957) and CHOLLET (1965). TUFTS—HANSEN (1933) and KOBEL (1954) could not prove metaxenie in pears.

Material and Method

Pollination studies were made between 1967 and 1970 at the Érd-Elvira Station of the Horticultural Research Institute with a variety collection grafted to wild pear stocks planted in 1953. Metaxenie studies were performed with ripe fruits obtained from the combinations of the pollinating partners, as shown in Table 1.

The great number of combinations of the pollen varieties as well as the similarly great number of ripe hybrid fruits in each combination (Tables 3 and 4) made it possible to perform extensive studies on the metaxenic effects occurring in pear varieties.

Among the pollinated varieties it is with the varieties Vilmos and Hardenpont that we demonstrate the metaxenic effects observed, since with these varieties a three year serie of data and high fruit number are available in the combinations examined.

The metaxenic effect most often manifests itself in changes in the size and shape of fruit, these questions are therefore the first to be discussed. Each year a different number of fruits were measured (Tables 3 and 4) with a slide-gauge, to two decimal places. Definitions of terms used for fruit measurements:

Fruit length: distance of two parallel tangential planes placed at the stem end and stylar end of the fruit respectively.

Largest fruit diameter: width of the largest cross section, distance of two parallel tangential planes placed at both sides of the fruit at right angles to the former planes.

Length of fruit neck: distance of the largest diameter from the fruit stem.

From the above measurements the following indices were determined:

Index of fruit shape: ratio of largest diameter to fruit length.

Index of the length of fruit neck: ratio of fruit neck length to fruit length.

The dispersion and standard deviation of the data obtained from the fruit measurements were also determined. The length of this paper does not make it possible to present these data; they can be found, however, in our earlier papers (NYÉKI 1970a, 1970b).

Table 1

Number of varieties and combinations included in the pollination studies, number of flowers pollinated and percentage of ripe fruits

Year	Number of varieties pollinated	Number of combinations of the pollen varieties	Number of flowers pollinated	Percentage of ripe fruits (maternal fertility of varieties)
1967	4	31	3.463	3.0
1968	24	240	26.208	1.9
1969	14	143	15.764	8.1
1970	26	231	28.018	6.4

Results

The size and shape of the fruit is a genetically determined characteristic of the variety. These features may vary between broad limits within the genotype, even in the case of identical pollinating varieties. Factors influencing them have been described by several authors in connection with pomiferous plants e.g. temperature, precipitation, root-stock, hormones, number and distribution of seeds in fruits, position of flowers in the crown and in the cluster, age and position of the generative parts, etc. Neither can the modifying effect of the above factors be completely eliminated when studying the metaxenic effects, and makes investigations into these questions very difficult.

We had to take the influence of the above modifying factors in consideration when studying metaxenic changes in pear varieties.

1. The range of variation in the pomometric values of fruits of a variety with in a variety collection, on the same tree. In a variety collection consisting of 410 varieties of the same age the pomometric response to cross pollination of a single tree in a selected variety was studied. The range of variation of

pomometric values was examined by crown levels and by the age of the generative parts; owing to lack of space, however, the results are summed up in Table 2.

The data of the table show a high degree dispersion of certain fruit characteristics, due presumably to different pollen varieties included in the collection.

Table 2

Range of variation in 1970 in the pomometric values of 1413 fruits grown ripe in one 17 year old Hardenpont pear tree grafted to a wild pear stock

Fruits			Shape index of fruits	Neck length index of fruits
length (mm)	diameter (mm)	neck length (mm)		
55→96	46.5→78	27.5→67.5	0.696→1.200	0.416→0.889

Table 3

Mean values of pomometric measurements made on ripe Vilmos hybrid fruits pollinated with various pollen varieties in 1968, 1969 and 1970

Pollen variety	Year	Number of fruits examined	Fruit length (mm)	Largest fruit diameter (mm)	Length of fruit neck (mm)	Shape index	Fruit neck index	Average number of fertile seeds per fruit
Bosc kobakja	1968	11	84.50	54.00	64.20	0.63	0.75	7.3
	1969	23	86.00	56.00	57.00	0.65	0.66	6.9
	1970	35	77.50	52.00	58.00	0.67	0.74	4.5
Pringalle vaj	1968	36	76.33	61.50	54.33	0.85	0.71	7.1
	1969	48	75.00	63.00	55.00	0.84	0.73	7.0
	1970	62	64.50	49.50	47.50	0.76	0.73	3.2
Dupuit asszony	1968	23	82.50	63.57	59.28	0.77	0.71	8.1
	1969	31	78.00	66.00	61.00	0.84	0.78	7.9
	1970	42	65.00	47.00	54.00	0.72	0.83	3.7

2. Changes in the pomometric values of fruits under the influence of definite pollen varieties, in various years. Results of investigations with the variety Vilmos are presented in Table 3, while those related to Hardenpont in Table 4.

The data of the tables show that irrespective of the year each pollinating variety has a tendency to have the same effect on the fruit characteristics of the pollinated varieties. This influence manifests itself in the length of the fruit, the largest diameter, the length of the fruit neck, and in the changes

Table 4

Mean values of pomometric measurements made on ripe Hardenpont hybrid fruits pollinated with various pollen varieties in 1968, 1969 and 1970

Pollen variety	Year	Number of fruits examined	Fruit length (mm)	Largest fruit diameter (mm)	Length of fruit neck (mm)	Fruit shape index	Fruit neck index	Average number of fertile seeds per fruit
Pringalle vaj	1968	17	86.90	72.40	57.00	0.83	0.65	3.0
	1969	41	85.00	76.00	59.50	0.89	0.70	2.9
	1970	38	74.00	65.50	49.70	0.89	0.66	3.0
Vilmos körte	1968	28	105.00	76.50	71.83	0.72	0.68	4.0
	1969	53	103.00	75.00	72.00	0.72	0.69	3.7
	1970	49	97.00	76.50	64.00	0.78	0.65	3.5
Clapp kedveltje	1968	14	77.50	65.23	55.60	0.84	0.71	3.5
	1969	29	74.50	60.50	50.00	0.81	0.67	3.0
	1970	37	73.50	66.00	48.00	0.89	0.65	2.9

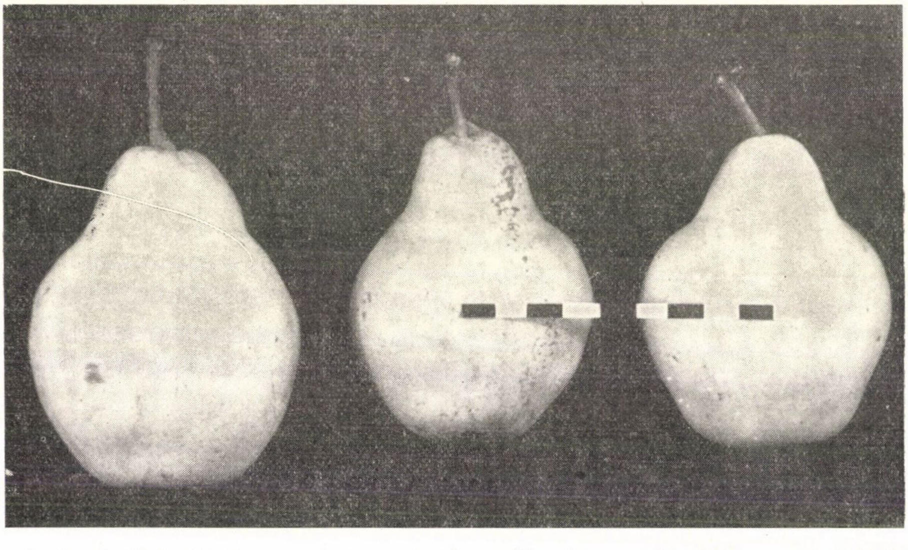

Fig. 1. On the left: Hardenpont fruit set by free pollination; on the right: two hybrid fruits of the Hardenpont variety pollinated with Vilmos pear

of the shape- and neck indices. Data show further, that the indices characterizing the morphological features of fruits are more constant than the numerical values obtained from the yearly measurements of fruit parts.

Metaxenic effects exerted on the size and shape of fruits are shown in Figs 1 and 2. In the combinations listed metaxenic changes could be observed soon after pollination, at an early stage of fruit development. Our data correspond to those obtained by Nagy (1957) in metaxenic studies performed with pear varieties, namely that the morphological characters of hybrid fruits vary from combination to combination. In certain cases these differences show the influence of the pollen variety. Each pollen variety has an influence of different extent on the various characteristics of the seed variety. The seed varieties show different degrees of resistance to the modifying effect of the pollen variety.

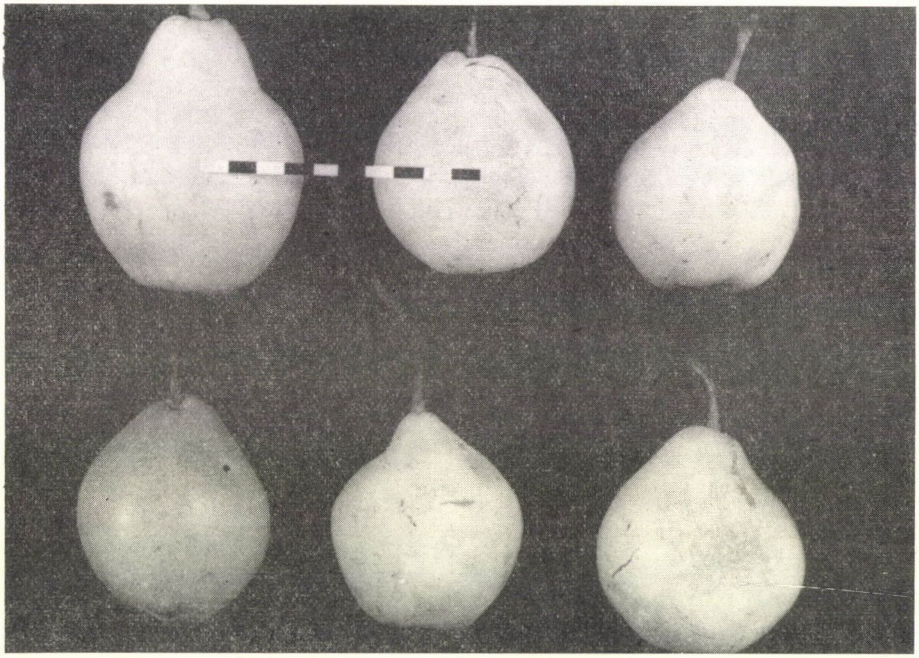

Fig. 2. In the left upper corner of the picture a Hardenpont pear originating from free pollination; the other five fruits are hybrid fruits of Hardenpont pollinated by Pringalle

3. Effect of pollen varieties on fruit colour and ripening time. Colour metaxenie was observed each year in the combination of Hardy (♀) ×Vilmos (♂). In the variety Hardy the lenticels on the hybrid fruits were of the same shape as in the variety Vilmos; in the combination of Hardenpont (♀)× Clapp kedveltje (♂) the colour of the epidermis on the hybrid fruits was quite similar to that in "Clapp kedveltje". As to the time of ripening, fruits in the combination of Hardenpont (♀)× Pringalle (♂) attained picking ripeness 5—7 days earlier each year than hybrid fruits in all the other combinations.

Conclusions

In contrast with KOBEL's (1954) opinion, numerous literary data as well as our own investigations suggest that in certain variety combinations producers and sellers must equally reckon with various (morphological and physiological) forms of metaxenie, as they may influence the commodity value of fruits.

Beyond a knowledge of the pollinating and fruit setting capacities of the varieties, optimum variety combinations in which the fruits of the main pollen varieties as far as possible have the same shape and colour as well as similar qualitative features with the other characteristics naturally differing — are very important for producers.

Further investigations are required to find out what the cause of the annual differences in the frequency of metaxenic changes in the individual variety combinations is.

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VARIATIONS IN PROTEIN AND LYSINE CONTENT IN SORGHUM VULGARE

By

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Ninety six jowar *(Sorghum vulgare)* materials comprising improved varieties, hybrids and locals were evaluated for the content of protein and lysine in the grain. With the values varying from 8.8% to 21.0% for protein and from 0.72% to 3.37% for lysine, these showed marked varietal differences for the above mentioned characters. Based on the protein content all were divided into low, medium and high protein groups. Sixty-three per cent of the materials came within the high protein group. The values for lysine varied from 1.24% to 3.37% with an average of 2.21% in low protein materials while in high protein materials the values varied from 0.72% to 2.95% with an average of 1.52%. The highly significant and negative correlation coefficient between the percentage of protein and the percentage of lysine (expressed on 100 g protein basis) showed that, in general, an increase in protein was associated with a decrease in the percentage of lysine. However, there were some lines such as IS 4532 and IS 4952 which had relatively higher lysine combined with higher protein content. This indicates possibilities of developing strains having high protein and high lysine by an extensive screening programme and adopting suitable breeding methods.

Introduction

Grain *Sorghum* (Jowar) occupies an important position in the cereal economy of India. Eighteen million hectares of land are under this crop and the annual production is about 9 million metric tonnes. It is consumed mostly by the rural population in certain regions in Andhra Pradesh, Mysore and Maharashtra where famines caused by the failure of crop owing to drought conditions are very frequent. Malnutrition arising out of calorie and protein deficiencies in the diet is prevalent among the people of these drought affected regions. Any nutritional programme aimed at improving the diet of these people should give due importance to the factor of personal preferences and also to the nutritional improvement of the food grains which form the bulk of the diet. This makes it urgent for the agricultural scientists to make concerted attempts to improve the nutritional quality in *Sorghum* which is generally known to be low in protein and lysine. Its higher leucine content is another disadvantage since an excess of this essential amino acid has been found to cause pellagra disease in human beings. In view of these nutritional inadequacies of the *Sorghum* grain, a programme for improving its nutritional quality has been recently initiated at the Indian Agricultural Research Institute. Varieties and hybrids developed at the Indian Agricultural Research Institute and other centres have been extensively tested for their quantity and quality of protein. Some of the results which are of importance to the breeders and nutritionists are reported here.

Material and Method

The grains of 96 varieties and hybrids grown uniformly under the co-ordinated *Sorghum* improvement programme were tested for their content of protein and lysine. The protein (N × 6.25) was determined according to the usual macro-Kjeldahl method. Lysine was determined by an automated colorimetric method using a Technicon automatic nitrogen analyzer. According to this method the determination of lysine in the hydrolyzate is achieved by a continuous colorimetric determination of CO_2, which is liberated on a mole per mole basis by the enzymatic decarboxylation of lysine using L-lysine decarboxylase. The method employed was basically the same as that described by SCHAIBERGER—FERRARI (1960), a modification introduced in the present estimations being the use of phosphate buffer of pH 6 instead of water for diluting the sample and the exclusion of the overflow sampler meant for providing small aliquots of the diluted sample.

Results

The results presented in Table 1 show that the values for protein varied from 8.8 to 21.0 per cent and those of lysine from 0.72 to 3.37 per cent. The varietal differences for these characters were very marked. Based on their protein content the various varieties and hybrids were divided into (I) low (less than 10% protein), (II) medium (10—12%) and (III) high (more than 12%) protein groups. Sixty-three per cent of the materials were in the high protein group while in the low protein group there were only 13 per cent. It may also be noted that 52 per cent of the materials in the high protein group had a protein content varying between 12 and 15% (Fig. 1). These results indicate that the protein level, in general, was higher in these varieties and hybrids.

The values for lysine varied from 1.24% to 3.37% with an average of 2.21% in low protein materials while in the high protein group the values varied from 0.72% to 2.95% with an average of 1.52%. In the medium group the values were 1.19% to 3.31% with an average of 1.80%. It is seen in Fig. 2 that 76% of the materials had a lysine content varying from 1 to 1.99%. In the low protein group 77% of the materials had a lysine content of more than 2% whereas in the medium group only 25% showed more than 2% lysine. In the high protein group the number of materials showing more than 2% lysine was still less, i.e. only 11%. MOHAN—DEOSTHALE (1969) reported that out of 332 *Sorghum* varieties tested, about 42 per cent had 10 to 12 per cent and 50 per cent had 1.5 to 2.0 per cent lysine.

Table 1

Variability for protein and lysine in Sorghum (Jowar)

Low protein type (Below 10%)

Variety/Hybrid	Protein	Lysine (g/100 g grain)	% Lysine (g lysine/100 g protein)
IS 1151	8.8	0.297	3.37
AKP-1	8.3	0.103	1.24
N-1	9.1	0.140	1.54
D-340	9.6	0.311	3.22
BH-4-1-4	9.1	0.226	2.48
Improved Saoner	7.8	0.178	2.28
RS 1	8.5	0.178	2.09
IS 4571	9.4	0.218	2.32
IS 5641	9.9	0.133	1.34
Medium protein type (10—12% protein)			
Co. 4	10.9	0.308	2.83
Co. 18	11.3	0.166	1.47
Co. 20	11.6	0.217	1.87
K. 2	10.8	0.200	1.85
Vzm-1	11.5	0.222	1.83
AKP-2	11.8	0.143	1.21
Nandyal	10.0	0.155	1.55
M-35-1 (Bijapur)	10.2	0.180	1.76
M-35-1 (Delhi)	10.4	0.124	1.19
PJ-4K	10.2	0.173	1.70
PJ-24K	10.0	0.173	1.73
NJ-156	10.0	0.129	1.29
Jowar No. 8	10.3	0.137	1.33
Satpani	10.1	0.160	1.58
Early 56	11.5	0.244	2.12
IS 4532	10.4	0.344	3.31
IS 4533	11.5	0.266	2.31
IS 4588	10.9	0.151	1.39
IS 4659	10.3	0.162	1.57
IS 4989	11.3	0.273	2.42
IS 4991	11.0	0.139	1.26
IS 5007	10.9	0.175	1.60
IS 5595	11.7	0.322	2.75
IS 5669	11.8	0.140	1.19

Table 1 (cont.)

Variety/Hybrid	Protein	Lysine (g/100 g grain)	% Lysine (g lysine/100 g protein)
High Protein (Above 12%)			
IS 84 (11)	14.4	0.103	0.72
IS 511	13.1	0.180	1.19
IS 815	15.3	0.208	1.36
IS 2031	15.1	0.182	1.21
IS 2944	15.1	0.234	1.55
IS 3796	13.4	0.194	1.45
IS 3797	14.2	0.160	1.13
IS 3924	13.0	0.143	1.10
IS 3922 (R-393)	12.6	0.171	1.36
IS 3922 (R-405)	13.2	0.176	1.33
IS 2954	13.3	0.222	1.67
R 473	14.4	0.279	1.94
IS 84	13.5	0.211	1.57
IS 3691	13.7	0.291	2.13
Kafir B	13.5	0.239	1.77
CSH 1	13.1	0.291	2.23
CSH 2	12.9	0.257	1.96
Co. 12	12.7	0.257	2.02
Co. 19	12.6	0.171	1.36
Co. 3	12.5	0.205	1.64
K. 3	14.3	0.205	1.43
N-6	13.2	0.167	1.27
N-10	13.0	0.215	1.65
Kalgonda	14.1	0.215	1.65
G. M. 1-5	14.0	0.195	1.39
Shenoli 4-2	12.4	0.140	1.13
C-10-2	13.1	0.155	1.18
B. P. 53	12.3	0.141	1.16
Vidisha 60-1	16.3	0.167	1.02
Ujjain 6	12.4	0.178	1.44
Fooder type small seeded			
Neel jowar	16.7	0.189	1.13
A-5-13-16	18.7	0.329	1.76
A-5-23-19	21.0	0.337	1.60
A-7-17-7	12.9	0.200	1.55
A-3-622-16	18.1	0.349	1.93
S-1-14-8	17.2	0.233	1.35

Variety/Hybrid	Protein	Lysine (g/100 g grain)	% Lysine (g lysine/100 g protein)
S-1049	15.7	0.233	1.45
IS 4520	13.4	0.260	1.94
IS 4525	12.2	0.162	1.33
IS 4526	13.8	0.218	1.58
IS 4624	13.8	0.173	1.25
IS 4645	12.4	0.204	1.65
IS 4911	13.4	0.138	1.03
IS 4952	14.7	0.434	2.95
IS 4953	12.6	0.278	2.21
IS 4960	15.2	0.173	1.14
IS 4961	13.8	0.252	1.83
IS 4987	14.8	0.196	1.32
IS 4988	14.1	0.145	1.53
IS 4990	12.8	0.317	2.48
IS 5006	12.2	0.137	1.12
IS 5039	14.2	0.230	1.62
IS 5542	14.2	0.153	1.08
IS 5566	12.6	0.167	1.33
IS 5591	14.3	0.171	1.20
IS 5596	14.6	0.244	1.67
IS 5604	14.7	0.233	1.58
IS 5609	12.6	0.289	2.29
IS 5614	14.2	0.171	1.20
IS 5615	13.3	0.184	1.38
IS 5644	14.9	0.155	1.04
IS 5665	12.9	0.140	1.09
IS 5813	13.9	0.155	1.12

It appears from the above results that as the protein content increased, the percentage of lysine (on the basis of protein) in general decreased. To confirm this point a scatter diagram between these two characters was drawn (Fig. 3). Total correlation between protein and lysine was next worked out for the 96 varieties. The correlation was equal to — 0.3613 — a value which is significant at 1% level showing thereby that an increase in protein is associated with a decrease in lysine. This relationship, however, is not linear as is seen from the low numerical value of the correlation and also by the wide scatter of the points. **Mohan—Deosthale** (1969) also reported a significant negative correlation between protein and lysine. It is worth pointing out

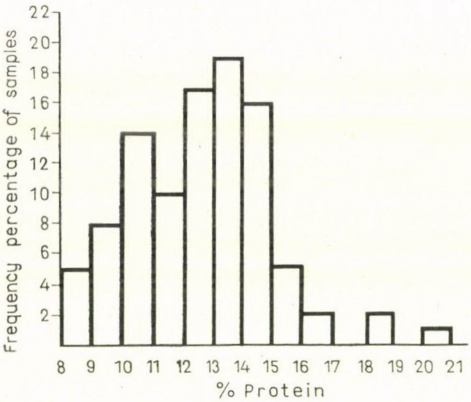

Fig. 1. Percentage frequency distribution of protein in Jowar varieties/hybrids (Based on 96 strains)

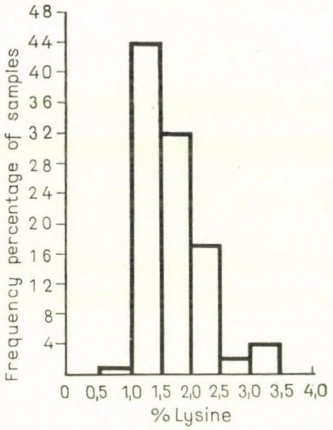

Fig. 2. Percentage frequency distribution of lysine in Jowar varieties/hybrids (Based on 96 strains)

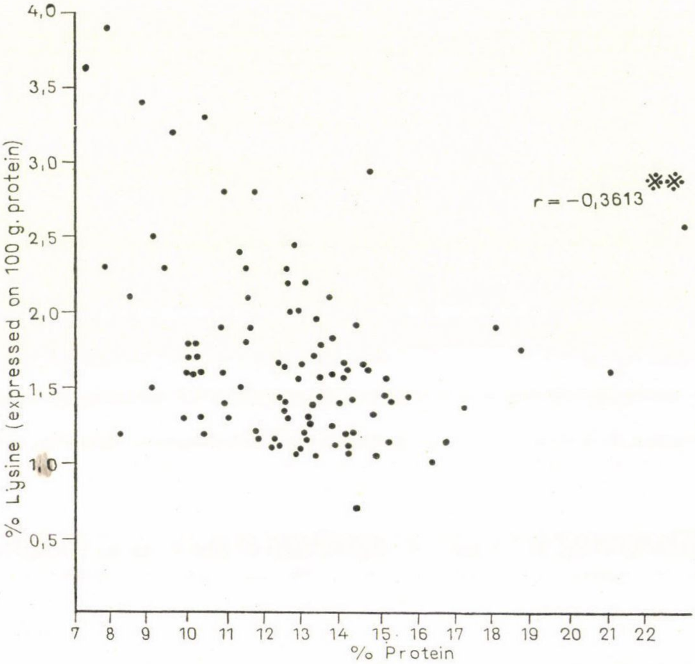

Fig. 3. Scatter diagram for percentage protein and percentage lysine (Based on 96 strains)

that IS 4532 in the medium and IS 4952 in the high protein groups showed higher lysine values of 3.22% and 2.95% respectively. These results suggest that by an extensive breeding and screening programme, it would be possible to select high protein-high lysine materials.

AKP-1, N-1 and IS 5641 in the low protein group were very low in their content of lysine. Varieties/hybrids such as Co.4, Early 56, IS 4532, IS 4533, IS 4989 and IS 5595 in the medium protein group and CSH.1, IS 3691, Co.12, IS 4952, IS 4953, IS 4990 and IS 5609 in the high protein group have come out promising for lysine.

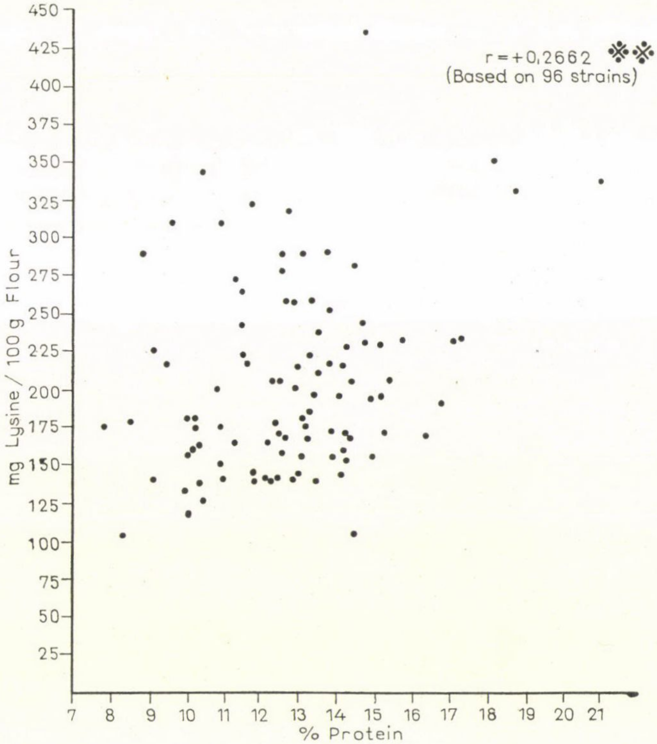

Fig. 4. **Scatter diagram of percentage protein and lysine (mg) expressed on 100 g flour basis (In Jowar)**

A similar scatter diagram (Fig. 4) for the percentage of protein and the absolute quantity of lysine expressed on 100 g flour basis was plotted. A highly significant and positive value of +0.2662 for the correlation coefficient was obtained. This shows that an increase in percentage protein was associated with an increase in the absolute quantity of lysine in the flour. Here also, this relationship, is not linear because of the wide scatter of the points and the low numerical value of the correlation. It may be noted that some of the strains such as CSH.2, R 473, IS 4525, IS 4961, A-5-13-16, A-5-23-19 and A-3-622-16, although they had less than 2% lysine on a protein basis, had on a flour basis a higher lysine content than that obtained in some of the higher lysine lines such as Early 56 and Co.12. This is obviously due to the higher protein content of the fairly average lysine content obtained in the former varieties/hybrids.

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STUDY ON THE PHYSIOLOGICAL AND BIOCHEMICAL BASES OF COMBINING ABILITY IN MAIZE LINES AND HYBRIDS

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By studying 20 biochemical indices in the generative organs of maize we tried to find correlations between quantities of the individual compounds and combining ability. In contrast with the supposed favourable effect of great differences in concentration we found that it was a definite level — up to about 10—20% — difference between the parents to be crossed that resulted in a favourable combination. The correlation is, however, only of tendential character, no direct parallelism can be proved. A positive correlation was found between the reproduction rate of Saccharomyces cultured in seed extract and the amount of yield as well as the good or bad combining ability of strains examined.

Introduction

With hybrids produced from inbred strains and lines a 20—30% yield increase has been achieved as compared to the former true-bred varieties. In order to gain hybrids the breeders produce, maintain and test thousands of strains every year. Testing reveals that not every line is able to increase viability in comparison with the control. Lines with good combining ability can be selected only by laborious experimentation, since selection has been performed empirically so far (**Pap** 1953, **Jugenheimer** 1958, **Brandolini** 1959). Out of the several thousand combinations only a low number is used in practice.

When evaluating the hybrid lines the researches assumed that the increased productivity was caused by the physiological differences of gametes (**Hagberg** 1953, **Győrffy** 1961, **Valyura** 1958). In order to characterize the latter chemical analyses of the generative organs were performed as well (**Polyakov** 1951, 1964, **Britikov** 1952, 1954, **Bálint** *et al.* 1958, 1960, **Gáspár** 1960).

In our experiments we aimed at producing known combinations as well as new ones. The generative organs of these combinations were analysed for differences and extent of change in the concentration of physiologically active matters. With these examinations we wished to throw light upon the prospective biological economic values, and through the differences arrive at — or at least approach — the causes of combining ability and over-development.

Materials and Methods

In the experiments six lines — C5, M14, WF9 American, O14, O118b ,156 Martonvásár — and four varieties — mindszentpusztai fehér Mf, Fleischmann korai Fk, martonvásári FB, Aranyözön Aö — were used.

With the method of top-crossing the following combinations were produced:
C5 × M14, C5 × WF9, C5 × O118b, C5 × 156, C5 × Fk, C5 × Aö, C5 × FB, C5 × Mf;
Mf × C5, Mf × M14, Mf × WF9, Mf × O118b, Mf × 156, Mf × Aö, Mf × FB, Mf × O14,
as well as the known standard hybrids: C5 × O14 and Mf × Fk.

The combining ability was determined by comparing the yields of the combinations with those of the known hybrids — C5 × O14, Mf × Fk — for three years at two sites. The comparative experiments were laid out in a 70 × 50 cm Latin square design, with 40 plants each in the four replications. Yield results were evaluated with the method of variance analysis.

In the course of analysing the generative organs of combinations we studied the first progamous phase of the process of fertilization — which depends on the character of interaction between pollen- and stigma tissues — further the end product, the seed — in which changes occur under the influence of the pollen, — and finally the possibility of approaching the problem of forecasting through biological methods.

The biochemical analysis of stigma- and pollen tissues as well as of the seed seemed to be most efficiently carried out through the comparison of the following indices: 1) Quantitative comparison of organic phosphorus compounds. Isolation was carried out with Ogur—Rosen's technique, destruction with the micro-Kjeldahl method, while phosphorus determination with that of Berenblum—Chain. 2) Trends were shown by the nucleic acid concentrations. Extraction was carried out with the Schneider—Schmidt—Tannhauser method, determination on the basis of ultra-violet absorption at a standard adenine level. Quantities were also checked by means of phosphorus- and sugar determination. Phosphorus was determined with Martyn—Doty's, while riboses with Brown's and Dische—Scheibert's (cit. Kovács 1958) methods respectively. 3) For the quantitative determination of vitamins the biotesting method was used. The total vitamin content was determined by Matskov's (1959) while the individual vitamins by Ogyintsova's (1959) method. 4) Protein determination was carried out with Nessler's reagent, after an acidic exposure.

Amino-acids were determined with chromatography (Hais—Macek 1961).

Results

Examination of the combining ability. Results obtained by a comparison between the yields of the combinations — Table 1 — show that there are differences in combining ability between lines and varieties used in the experiments.

In case of lines crossed two, while with varieties crossed eight combinations exceeded the standard hybrid.

Examination of generative organs. Concentrations of the biochemical indices of pollen- and stigma tissues are shown in Tables 2 and 3. The tables present data on the generative organs of the parents of the standard hybrid, as well as of parents showing better and poorer combining ability respectively than the standard. The analysis disclosed that concentrations of indices examined are different both between the various organs and within them. The character of differences between the organs seems to indicate that the various concentrations are complementary to each other according to their respective biological role (pollen- and stigma values of parents). Differences within the organs suggest that generative organs of parents involved in the combination

Table 1

Yield results of combinations

Combinations	Dry yield F_1	Dry yield F_2	Combinations	Dry yield F_1	Dry yield F_2
	kg/plot			kg/plot	
C5 × M14	10.29	9.37	Mf × M14	9.25	7.52
C5 × 0118 b	9.90	6.74	Mf × FB	9.11	6.74
C5 × 014 st	8.76	6.74	Mf × 0118 b	8.56	7.52
C5 × Aö	8.93	6.92	Mf × WF9	8.41	7.26
C5 × FK	8.53	7.35	Mf × Aö	8.32	7.00
C5 × WF9	8.14	6.13	Mf × C5	8.05	7.35
C5 × 156	8.15	7.73	Mf × 014	7.62	6.13
C5 × FB	7.79	7.44	Mf × Fk st	7.46	5.87
C5 × Mf	7.78	7.70	Mf × 156	7.35	6.92
S.d. 5%	1.32	1.92	S.d. 5%	1.24	1.75

possess divergent properties. This biochemical heterogeneity means the realization of existing differences in the genotype. Data show further, that concentrations in the various organs are equally different when generative organs of plants in the inbred line are analysed, when pollen- and stigma tissues of F_1 plants are examined or corresponding organs of plants in varieties maintained by intercrossing compared. Data give information on the different concentrations in the generative organs of tester plants too, which originate from their genetic differences. (See C5 in Table 2 and data of Mf in Table 3). Literary data as well as research work of similar nature carried on at the department have disclosed that the normal course of the sexual process is preconditioned by a difference in the biologically active materials of pollen- and stigma tissues.

Further investigations called attention to the necessity of expounding this finding from the point of view of combining ability. A comparison of the indices examined revealed that a general difference is not enough as a precondition. From the point of view of a positive interaction between materials existing or developing "corresponding" differences are necessary. Only a definite level difference may lead to the balance that ensures the first progamous phase of the process. In the case of self-pollination preconditions of the realization of "corresponding" differences seem to be absent, only cross-pollination — in present case C5 and 014, C5 and 0118b, — as well as line crossing of M14 — and crossing of varieties Mf and FB make it possible.

Table 2

Concentrations of generative organs in lines,

Biochemical indices examined	C5		O14	
	pollen	stigma	pollen	stigma
RNA*	0.0842	0.1544	0.1174	0.2592
Inosite**	220.00	314.00	540.00	495.94
DNA*	0.0278	0.0472	0.0236	0.0470
Alcohol soluble-P***	91.2	96.6	74.5	74.1
$B_1 + B_6$**	93.05	12.86	59.75	30.37
Acid soluble-P***	63.2	38.2	46.8	36.4
Nicotine acid**	22.0	7.64	28.85	9.97
Panthotene acid**	25.50	4.04	25.70	10.47
Nucleid acid-P***	16.6	11.5	14.5	27.1
Lipoid soluble-P***	14.7	9.8	21.7	7.2
Protein per cent	18.9	10.9	14.8	10.9
Biotine**	3.98	1.2	4.85	1.44

Biochemical indices examined	C5×O14		C5×O118 b	
	pollen	stigma	pollen	stigma
RNA*	0.0782	0.4314	0.1373	0.2453
Inosite**	600.0	583.0	1050.0	550.0
DNA*	0.0229	0.0554	0.0175	0.0243
Alcohol soluble-P***	55.9	51.7	53.5	68.9
$B_1 + B_6$**	105.84	20.37	63.12	22.50
Acid soluble-P***	30.7	9.9	33.4	8.2
Nicotine acid**	51.6	10.40	51.20	10.70
Panthotene acid**	30.84	11.50	34.68	11.0
Nucleic acid-P***	8.3	26.8	6.5	18.3
Lipoid soluble-P***	3.2	10.3	4.8	15.7
Protein %	17.0	12.0	16.1	10.9
Biotine**	7.92	2.70	9.12	2.80

Note: * = adenine μM/g · dry matter
** = μg/g · dry matter
*** = μg P/g · dry matter

Analysis of correlation between biochemical indices examined and productivity pointed out a tendency-like rather than direct correlation between indices examined and combining ability. The tendential character means that there are now larger, now smaller differences in the concentration of materials *varieties and F_1 plants of different combining ability*

O118 b		FB		S.d. 5%	
pollen	stigma	pollen	stigma	pollen	stigma
0.1230	0.1320	0.1310	0.1566	0.0490	0.0904
625.40	563.74	563.80	432.50	11.10	22.4
0.0200	0.0190	0.0174	0.0304	0.0091	0.0045
64.2	90.6	80.3	57.3	3.8	11.3
61.35	33.99	80.30	18.0	11.1	22.4
41.0	40.4	34.2	22.4	2.5	3.2
29.05	11.17	34.35	9.0	1.1	2.4
27.90	4.93	26.15	4.73	1.1	2.4
13.7	18.8	10.4	6.7	2.1	4.5
18.1	9.9	5.5	11.6	1.7	2.5
16.1	11.9	16.3	13.0	0.95	1.2
3.36	1.44	3.98	1.32	1.1	2.4

C5×FB		S.d. 5%	
pollen	stigma	pollen	stigma
0.1343	0.1912	0.0545	0.0964
1200.0	316.0	11.1	22.4
0.0234	0.0262	0.0094	0.0055
47.2	115.0	4.2	14.2
48.72	15.25	11.1	22.4
37.3	22.6	2.8	5.2
43.20	6.90	1.1	2.4
27.28	3.46	1.1	2.4
7.6	13.5	2.7	5.8
3.2	12.2	1.5	2.5
15.2	9.5	0.95	1.2
4.8	1.7	1.10	2.4

examined between the organs, and that differences always result in a yield increase (C5 × M14, C5 × O118b). This suggests that in the various combinations the high yields of progenies are produced in different ways depending on the parents to be crossed. The combining ability of maize as judged by the amount of yield is a complex system including the adequate proportions of genetic material (DNA, RNA) as well as developing of an improved system of physiologically active materials. The adequate proportion of the genetic material,

Table 3

Concentrations of generative organs in varieties,

Biochemical indices examined	Mf		Fk	
	pollen	stigma	pollen	stigma
RNA*	0.1332	0.2456	0.1146	0.1688
Inosite**	662.20	364.0	406.50	540.00
DNA*	0.0202	0.0428	0.0124	0.0272
Alcohol soluble-P***	72.9	60.8	65.2	64.5
$B_1 + B_6$**	94.15	13.62	54.90	14.30
Acid soluble-P***	53.2	45.4	45.6	39.5
Nicotine acid**	24.75	7.64	22.50	9.04
Panthotene acid**	26.3	4.04	27.0	4.04
Nucleid acid-P***	10.3	32.9	13.5	8.1
Lipoid soluble-P***	20.1	6.7	14.4	7.1
Protein %	16.0	11.0	16.3	15.3
Biotine**	5.06	1.44	3.90	1.32

Biochemical indices examined	Mf×Fk		Mf×FB	
	pollen	stigma	pollen	stigma
RNA*	0.0626	0.2902	0.1438	0.2504
Inosite**	1575.0	370.0	975.0	375.0
DNA*	0.0117	0.0290	0.0215	0.0373
Alcohol soluble-P***	45.9	108.2	45.8	47.1
$B_1 + B_6$**	50.52	10.12	32.16	15.02
Acid soluble-P***	29.2	6.9	35.3	13.8
Nicotine acid**	36.60	6.04	40.44	8.00
Panthotene acid**	22.44	3.08	26.16	3.78
Nucleid acid-P***	9.7	18.3	6.4	4.3
Lipoid soluble-P***	3.4	12.2	2.8	4.6
Protein %	—	—	18.1	11.9
Biotine**	5.04	1.8	5.52	1.96

Note: * = adenine μ M/g · dry matter
** = μg/g · dry matter
*** = μg P/g · dry matter

the free combination, the development of more perfect and balanced physiological systems are influenced — in addition to the genetic basis — by internal and external factors as well. Consequently, studies of any index or character present only tendency-like relations even when gametes can be directly *lines and F_1 plants of different combining ability*

FB		O14		S.d. 5%	
pollen	stigma	pollen	stigma	pollen	stigma
0.1310	0.1576	0.1174	0.2592	0.0490	0.0904
363.80	432.50	540.00	495.94	11.1	22.4
0.0174	0.0304	0.0236	0.0470	0.0091	0.0045
80.3	57.3	74.5	74.1	3.8	11.3
80.3	18.0	59.75	30.37	11.1	22.4
34.2	22.4	46.8	36.4	2.5	3.2
34.35	9.0	28.85	9.97	1.1	2.4
26.15	4.73	25.7	10.47	1.1	2.4
10.4	6.7	14.5	27.1	2.1	4.5
5.5	11.6	21.7	7.2	1.7	2.5
16.3	13.0	14.8	10.9	0.95	1.2
3.98	1.32	4.85	1.44	1.1	2.4

Mf×O14		S.d. 5%	
pollen	stigma	pollen	stigma
0.1263	0.2995	0.0510	0.0928
660.0	435.5	11.1	12.4
0.0296	0.0225	0.0106	0.0067
65.3	44.3	3.9	13.5
92.64	19.12	11.1	22.4
29.8	16.4	2.6	4.4
60.0	8.64	1.1	2.45
28.20	10.74	1.1	2.45
9.6	17.2	2.4	6,7
4.7	17.9	1.9	2.8
15.2	11.0	0.9	1.2
5.64	2.30	1.1	2.4

analysed. Data show first of all that hybrids with good combining ability can be produced by crossing high quality lines, if besides good phenotype characters the proportions of active matters do not show too great differences. A high extent of accumulation may upset the equilibrium (as shown by the examination of the generative organs of F_1 plants of which results are shown in the lower part of Tables 2 and 3), disturb the cell metabolism, or else, vegetative development will be too intensive and nutrient supply in the generative or-

Table 4

Concentrations of seeds in lines,

Biochemical indices examined	C5	C5×O14	O14
RNA*	0.1734	0.1564	0.2130
Inosite**	45.81	44.70	58.32
DNA*	0.0495	0.0583	0.0652
Alcohol soluble-P***	40.5	84.4	73.0
$B_1 + B_6$**	5.85	6.08	6.41
Acid soluble-P***	82.2	76.8	27.3
Nicotine acid**	1.97	2.89	2.52
Panthotene acid**	1.26	1.80	1.60
Nucleic acid-P***	38.0	43.9	50.2
Lipoid soluble-P***	47.0	37.4	34.3
Protein %	11.9	10.0	9.0
Biotine**	0.039	0.045	0.057

Biochemical indices examined	Mf	Mf×Fk	Fk
RNA*	0.0534	0.1015	0.1600
Inosite**	32.82	106.0	87.51
DNA*	0.0170	0.0395	0.0466
Alcohol soluble-P***	140.2	47.8	87.2
$B_1 + B_6$**	7.05	11.25	9.37
Acid soluble-P***	53.2	72.7	52.3
Nicotine acid**	2.50	1.70	1.51
Panthotene acid**	1.69	3.44	3.32
NA-P***	27.0	26.2	41.5
Lipoid soluble-P***	39.8	19.1	57.3
Protein %	11.0	12.1	11.3
Biotine**	0.042	0.170	0.120

Note: * = adenine μM/g · dry matter
** = μg/g · dry matter
*** = μg P/g · dry matter

gans insufficient. In this case the vegetative development of F_1 plants shows heterosis, which cannot, however, be found in the grain yield where a decrease is observed instead. We found, further, that by inbreeding, biochemical properties characteristic of the line become more easily demonstrable. Thus, *varieties and hybrids of different combining ability*

C5×O118b	O118b	C5×FB	FB	S.d. 5%
0.2129	0.1946	0.1128	0.0676	0.0540
23.6	47.91	28.0	83.34	12.1
0.0381	0.0268	0.0578	0.0226	0.0210
109.9	88.5	146.6	143.3	28.5
4.50	8.1	6.0	4.93	2.1
73.4	74.5	92.3	63.2	9.1
1.54	3.38	2.7	2.50	0.6
1.26	1.47	1.5	1.28	0.6
52.1	45.3	32.9	19.3	12.1
33.4	31.8	25.5	38.2	12.4
10.2	11.1	9.4	11.8	1.15
0.034	0.054	0.015	0.041	0.02

Mf×FB	FB	Mf×O14	O14	S.d. 5%
0.1466	0.0676	0.2439	0.2130	0.0540
120.8	83.34	28.9	58.32	12.1
0.0606	0.0226	0.0846	0.0652	0.0210
103.5	143.4	159.3	73.0	28.5
5.58	4.93	4.50	6.41	2.1
67.4	63.2	61.5	27.3	9.1
3.38	2.50	2.57	2.52	0.8
1.70	1.28	1.66	1.60	0.6
38.8	19.3	42.4	50.2	12.1
20.0	38.2	46.2	34.3	12.4
12.1	11.8	10.3	9.0	1.15
0.058	0.041	0.036	0.057	0.02

direction, extent and effect on the progeny of changes can be better followed. From this point of view variety is not a good object of examination.

Biochemical examination of seeds. Biochemical indices of seeds are included in Table 4. The analysis of seeds disclosed well established differences between lines and varieties. Changes of materials accumulating in the seed in the case of crossing can be measured. In the case of line crossing the amount of phosphorus compounds increased while that of the vitamins decreased. In the case of varieties crossed it happened the other way round. Our results agree with literary data on the different degrees of accumulation of phosphorus compounds, nucleic acids, vitamins, etc. in the hybrid seeds. These data were considered by the authors as representing favourable characteristics or abilities from the point of view of hybrid progenies. It was in essentials on these results that Matskov based his heterosis "prognosis" method. CHERRY *et al.* (1961) consider the concentration of RNA, the ratio of ADP/ATP as an index of heterosis. The authors carried out investigations with several combinations. When, however, the number of combinations to be examined is raised and seeds of not only inbred hybrids analysed, results are not unequivocal. Analysis of the examination results of seeds has shown that a high degree accumulation cannot unanimously be considered a favourable characteristic. High extent changes — either in the form of reduction or in that of accumulation — result in yield decreases of similar extent. Comparison between seed analysis results and yield results called attention to the fact that only a low extent change affecting but one or the other compound thus promoting an equilibrium is favourable from the point of view of the progeny. That is why we emphasize the tendential character of he correlation.

Examination of a possibility of forecasting. When studying the biological method of a possible forecasting we started from Matskov's results, though we examined seeds instead of leaves. After several biotests applied our results showed that reproduction intensity of Saccharomyces cerevisiae followed the changes of the yield amounts rather reliably.

We have already mentioned that hybrids exceed in most cases the examined characteristics of the parents, therefore combining ability should be evaluated — as in the breeding practice — and possibilities of forecasting reckoned in comparison to a hybrid known for its good combining ability. Table 5 expresses the amount of yield in the percentage of standard hybrids. The reproduction value of the fungus is similarly expressed as a per cent of the reproduction value of the standard hybrid. Within a limit error percentage values agree in case of both yield and fungus reproduction, when either the parents or the hybrids are compared. With variety hybrids the percentage value of fungus reproduction is higher than that of the yield. This is connected with our earlier mentioned result, namely that the quantity of vitamins increases when varieties are crossed. With the Saccharomyces cerevisiae the total amount of the so called bio-materials is determined. This fungus is suitable for serial examinations. Lines to be crossed are many-sidedly studied by the breeders. An extension of these studies by means of the biological method is recommended, since it would help in making a final decision on partners used in a combination. Our data pointed out, further, that joint determination of materials in the bio-group is not sufficient. Interactions of materials as well as their adequate ratios should also be studied. They should be determined simultaneously with other biotests or methods. Saccharomyces-less strains are considered to be suitable for this purpose. Data of the two kinds of simultaneous determination confirmed the existence of a positive correlation between the reproduction of Saccharomyces and the good or bad combining ability of strains examined.

Table 5

Comparison between the yield amount and the reproduction of Saccharomyces cerevisiae on the basis of their percentage values

Index examined	C5	C5×O14	O14	C5×O118b	O118b	C5×FB	FB	S.d. 5%
Grain yield kg	4.59	8.76	4.85	9.90	5.68	7.79	8.76	1.32
Grain yield as a % to the st hybrid	52	100	68	111	64	80	100	
Reproduction of Sacch. c. as a % to the st hybrid	55	100	63	111	70	92	97	
	Mf	Mf×FK	FK	Mf×FB	FB	Mf×O14	O14	S.d. 5%
Grain yield kg	7.6	7.46	8.81	9.11	8.76	7.62	4.85	1.24
Grain yield as a % to the st hybrid	101	100	118	122	117	102	55	7
Reproduction of Sacch. c. as a % to the st hybrid	125	100	84	134	97	102	63	11

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EFFECT OF SOME BIOACTIVE COMPOUNDS ON NITROGEN METABOLISM IN THE MYCELIUM OF AGARICUS BISPORUS MÖLL. ET SCHÄFF. AND COPRINUS COMATUS FR.

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The authors studied the effects of several groups of bioactive substances (cytokinins, auxins, gibberellins, growth retardants and senescing agents) on the mycelium development, dry matter content, total nitrogen and protein nitrogen content of the fungi: *A. bisporus* Möll. et Schäff. and *C. comatus* Fr. belonging to *Basidiomycetes.* High biological effectivity was found in the case of purin and pyrimidin analogues as well as under the influence of treatments with morphactines, B-9 and Ethrel; the latter induces senescence in higher plants. In the case of excitation a concentration dependent effectivity could also be demonstrated. The selectivity of coffein and phenobarbituric acid is worth being emphasized as causing an increase of dry matter in *A. bisporus* Möll. et Schäff. and a specific inhibition in *C. comatus* Fr.

Introduction

Miller (1967) isolated endogenous cytokinins from the mycelium of mycorrhizal fungi, and indicated zeatin and zeatin-riboside occurring during this process. In our earlier experiments cytokinin-activity was demonstrated in phytopathogenic fungi, especially in the phase of sporulation, but a considerable amount of endogenous cytokinin-like compounds also get into the tissues of the host organism through the killed off sterile hyphae (Király—Pozsár—El Hammady 1966, Király—El Hammady—Pozsár 1967). On the other hand, the biological activity of endogenous cytokinins isolated from the fruiting bodies of macro fungi was compared with the endogenous factors of higher green plants (Szabó—Pozsár—Kota 1970), with the surprising result that the cytokinin activity was nearly ten times as high in the fruiting body of the examined macro fungi as in the leaves of papilionaceous plants, although it is not identical with kinetin or benzyladenine.

Ferenczy—Stefandel (1958) studied the action of the growth regulator auxin and found that auxins exercised a higher or lower growth inhibiting effect on the *Aspergillus niger* cultures examined.

On the above basis thorough studies were made on the effect of numerous synthetic cytokinins — and hormone-like growth regulators in higher green plants as well as that of antimetabolites — on growth, dry matter accumulation and nitrogen metabolism in the mycelium of two macro fungi — *A. bisporus* Möll. et Schäff. and *C. comatus* Fr. — belonging to the class of *Basidiomycetes.*

Material and Method

The culture stocks of *A. bisporus* Möll. et Schäff. and *C. comatus* Fr. were made available by Bohus (Botanical Collection of the Museum of Natural Science, Budapest) for which we — here too — express our thanks.

The synthetic basic culture medium was prepared taking in consideration the ion concentrations found optimum by TRESCHOW (1944) for *A. bisporus* Möll. et Schäff. mycelium cultures according to the following:

compounds:	g
D-glucose	20.0
Glycine	1.0
K_2HPO_4	0.5
KH_2PO_4	0.5
$CaCl_2 \cdot 2H_2O$	0.2
$MgSO_4 \cdot 7H_2O$	0.1
$FeSO_4 \cdot 7H_2O$	0.01
H_3BO_3	0.01
$CuSO_4 \cdot 5H_2O$	0.005
$MnSO_4 \cdot H_2O$	0.001
$ZnSO_4$	0.001
Thiamine	$2 \cdot 10^{-3}$
Biotin	10^{-5}
Distilled water up to	1000
pH 7.0	

Considering the fact that among the fungi examined it was *A. bisporus* Möll. et Schäff. which had higher nutrient requirements, in the interest of comparability the above described liquid culture medium was used for the other (in the present case *C. comatus* Fr.) fungi too.

The active agents were added to the culture medium at concentrations of 1, 10 and 100 ppm, then from the culture medium containing the active agents 10 ml was placed in each normal test tube of which 4 replications were prepared per treatment and fungus species.

The culture media were sterilized in an autoclave for 20 minutes in steam at an overpressure of 1 atmosphere.

The stock cultures were maintained in a refrigerator at +4°C, on a basic culture medium completed with 2 per cent agar-agar. When inoculating, in the case of *C. comatus* Fr. culture medium free aerial mycelium was used as inoculum, while in the case of *A. bisporus* Möll. et Schäff. a piece of the size of 1 mm^2 was taken from a layer of the culture medium interlaced with mycelium. The inoculated cultures were incubated in a thermostat at a temperature of 25°C. During the period of incubation the growth of the cultures was compared with that of the control every third day on the basis of macroscopic observations, then on the 21st day after inoculation the colonies were separated from the culture media by filtration and washing with sterile water.

In addition to the dry matter content the total nitrogen and the insoluble protein nitrogen in 10 per cent TCA at +4°C temperature were determined by the micro-Kjeldahl method and expressed as a percentage of dry matter content.

The active agents examined belong to the following groups: 1. cytokinins: purin analogues (kinetin, benzyladenine, coffein, theobromine, theophylline), pyrimidine analogues (6-methyl-uracil, 4-methyl-2-thio-uracil, barbituric acid, pheno-barbituric acid, thio-barbituric acid), imidazoles (benzimidazole, 5,6-dimethyl-benzimidazole); 2. auxins: indoleacetic acid, 2,4-dichloro-phenoxi acetic acid; 3. gibberellins: gibberellic acid (GA_3), morphactine (2-chloro-9-hydroxy-fluoren-9-carbon acid methyl esther); 4. growth inhibitors: chloro-cholin-chloride (CCC), succinic acid-dimethyl-hydrazide (B-9); senescence inducing substance: 2-chloro-ethyl-phosphoric acid (Ethrel).

Results

Purin and pyrimidin analogues of 1, 10 and 100 ppm concentration (Table 1), as well as auxins, gibberellins and other growth regulators (Table 2) have a specific and varied effect on the weight and dry matter content of *A. bisporus* Möll. et Schäff. and *C. comatus* Fr. mycelium cultures.

Table 1

Effect of purin and pyrimidine analogues at 1, 10 and 100 ppm concentration on colony weight and dry matter content in the mycelium cultures of A. bisporus Möll. et Schäff. and C. comatus Fr.

Treatment	Cc. in ppm	Average colony weight (mg)		Dry matter (mg)		Percentage proportion of dry matter related to control	
		Agaricus	*Coprinus*	*Agaricus*	*Coprinus*	*Agaricus*	*Coprinus*
Control		290.4	152.4	7.17	6.69	100	100
Kinetin	1	319.9	170.5	11.80	11.85	164	177
	10	362.1	142.8	12.13	8.04	169	120
	100	94.5	6.0	5.09	—	71	—
Benzyladenine	1	366.2	208.3	10.14	15.84	141	236
	10	305.0	121.8	7.75	6.70	108	100
	100	33.6	0.0	—	0.0	—	0
Coffein	1	244.2	107.8	7.59	5.73	105	85
	10	309.0	94.8	13.32	5.05	184	75
	100	225.8	78.1	10.72	3.82	149	57
Theobromine	1	256.5	195.9	12.11	9.27	168	138
	10	179.1	153.1	11.66	11.27	162	168
	100	266.1	112.2	11.42	6.93	159	103
Theophylline	1	233.8	213.5	6.78	12.28	94	183
	10	249.9	146.9	10.69	11.30	149	169
	100	204.8	164.0	9.60	10.63	134	159
Barbituric acid	1	279.5	180.0	12.17	9.79	169	146
	10	272.3	237.0	12.44	13.08	173	195
	100	332.9	189.0	13.55	11.34	189	169
Thiobarbituric acid	1	338.2	270.5	12.48	12.93	174	193
	10	272.1	211.3	13.47	8.35	188	124
	100	191.4	128.0	6.01	6.63	83	99
Phenobarbituric acid	1	320.1	157.7	14.47	5.30	201	79
	10	311.3	124.9	12.14	4.62	169	69
	100	181.0	74.9	9.32	3.20	130	47
6-methyl-uracil	1	209.3	254.8	8.79	13.76	122	205
	10	251.7	222.3	10.02	12.00	139	179
	100	252.2	204.4	13.34	11.16	186	166
4-methyl-2-thiouracil	1	247.5	255.4	15.27	13.91	213	208
	10	296.7	260.3	13.29	12.28	185	183
	100	156.1	255.1	9.58	10.87	133	162

Table 2

Effect of benzimidazoles, auxins, gibberellins and other growth substances at 1, 10 and 100 ppm concentration on colony weight and dry matter content in mycelium cultures of A. bisporus Möll. et Schäff. and C. comatus Fr.

Treatment	Cc. in ppm	Average colony weight (mg)		Dry matter (mg)		Percentage proportion of dry matter related to control	
		Agaricus	*Coprinus*	*Agaricus*	*Coprinus*	*Agaricus*	*Coprinus*
Control		290.4	152.4	7.17	6.69	100	100
Benzimidazole	1	254.6	156.4	6.56	8.20	91	122
	10	161.9	120.9	4.30	6.67	59	99
	100	86.9	58.2	1.31	4.57	18	68
5,6-dimethyl-benzimidazole	1	125.4	135.3	3.90	6.10	54	91
	10	118.8	76.8	3.62	5.32	50	79
	100	70.0	25.9	0.72	2.69	10	40
IAA	1	273.7	234.2	15.49	12.22	216	182
	10	272.7	181.9	14.29	10.86	199	162
	100	0.0	22.8	0.00	—	0	—
2,4-D	1	258.7	204.2	9.60	11.09	134	165
	10	160.8	181.1	9.50	10.74	132	160
	100	137.3	120.9	8.64	7.62	120	114
GA_3	1	239.0	98.4	11.15	5.72	155	85
	10	287.5	179.7	12.14	8.36	169	125
	100	310.4	98.5	13.84	5.52	193	82
2-chlorofluorenol	1	311.1	144.0	12.94	8.74	180	130
	10	110.1	146.0	10.74	8.64	149	129
	100	0.0	0.0	0.00	0.00	0	0
CCC	10	106.9	182.7	5.95	8.44	83	126
	100	170.8	131.4	8.88	5.23	123	78
B-9	1	207.7	163.3	10.78	7.36	150	110
	10	237.0	131.7	12.87	5.44	179	81
	100	185.5	132.0	7.98	5.40	111	80
Ethrel	1	258.5	272.7	13.34	11.50	186	172
	10	277.1	372.6	14.41	14.64	201	218
	100	299.9	155.9	12.03	5.05	167	75

Apart from the stimulating effect of 1 and 10 ppm kinetin and benzyladenin, the purin type alkaloids (theobromine, theophylline, coffein) have a remarkably positive effect too, mostly even at a concentration of 100 ppm, and so have the pyrimidine compounds of barbituric acids and their derivatives (barbituric acid, pheno-barbituric acid, thio-barbituric acid). Accumulation of dry matter is further stimulated by the cytokinin-like pyrimidine-analogue, 6-methyl-uracil, and by the antimetabolite-type 4-methyl-2-thiouracil too.

The other compounds are of positive effect, too — mainly at concentrations of 10 and 1 ppm— especially the 10 ppm Ethrel.

Table 3

Percentage stimulation of total nitrogen content by the biological active substances examined, in A. bisporus Möll. et Schäff. cultures

Active substances	Concentration in ppm	Dry matter (mg)	Percentage of total nitrogen in dry matter	Stimulation percentage related to total nitrogen
Control	—	7.17	4.35	0
4-methyl-2-thiouracil	100	9.58	7.73	78
CCC	10	5.95	5.63	29
IAA	10	14.29	5.03	16
2,4-D	1	9.60	4.79	10
GA_3	100	13.84	4.68	8
Theophylline	1	6.78	4.56	5
2-chloro-ethylphosphoric acid	100	12.03	4.41	1

Colony growth is decidedly inhibited by the 100 ppm kinetin, benzyladenine and benzimidazole, by dimethyl-benzimidazole at all concentrations examined, by 100 ppm thiobarbituric acid, 100 ppm indoleacetic acid, as well as by the 100 ppm 9-chloro-fluorenole derivative.

It is worth emphasizing that coffein and pheno-barbituric acid had a selective effect, namely they stimulated a dry matter increase in *A. bisporus* Möll. et Schäff., while an inhibition considered specific, was found in the case of *C. comatus* Fr.

Results showing that plant growth regulators increased the total nitrogen content expressed as a percentage of the dry matter content, compared with the control are presented in Table 3. The uracil derivative, CCC and auxins showed a specific stimulation.

Many compounds increased the proportion of protein nitrogen (Table 4). The stimulating effect was, however, mostly of a low level. In the case of theophylline all three concentrations examined showed outstanding effects. The 6-methyl-uracil was positive at 100 ppm, indoleacetic acid at 10 ppm and gibberellic acid especially at 100 ppm.

The investigations show that the growth regulators of green plants are not ineffective in influencing the growth and nitrogen metabolism of fungi belonging to a different taxonomic category, but it must by all means be emphasized that the efficient concentrations are at a much lower scale of values than in the case of green leaf and shoot tests or in vitro tissue and organ cultures. It is possible that the hormone translocation in the tissues of higher plants is of a lower extent, and the lower biological effectivity is due to the higher immobility.

Table 4

Compounds increasing the percentage of protein nitrogen, with the favourable concentration, the protein nitrogen percentage and the stimulation percentage as compared to the control presented, in A. bisporus Möll. et Schäff.

Active substances	Concentration (ppm)	Percentage proportion of total nitrogen to dry matter	Percentage proportion of protein nitrogen to total nitrogen	Stimulation percentage as compared to the control
Control	—	4.35	64.3	—
Kinetin	1	2.61	65.0	1.1
Benzyladenine	10	3.78	65.8	2.2
Theobromine	1	2.52	65.7	2.2
Theophylline	10	2.50	67.2	4.5
Theophylline	100	3.35	71.3	10.8
Dimethylbenzimidazole	10	3.44	64.9	0.9
Thiobarbituric acid	100	3.47	65.3	1.4
Phenobarbituric acid	1	1.85	65.7	2.1
Phenobarbituric acid	10	2.89	67.0	4.2
6-methyl-uracil	10	1.87	65.3	1.6
6-methyl-uracil	100	0.78	68.9—	7.0
4-methyl-2-thiouracil	100	7.73	66.4	3.2
IAA	10	5.03	69.4	7.8
GA_3	100	4.68	65.9	18.0
2,4-D	10	2.81	68.4	6.3
2,4-D	100	3.35	66.4	3.3
2-chloro-fluorenol	10	2.17	68.6	6.6

In the course of our investigations we arrived at the unexpected conclusion that, on the one hand, the natural plant substances of the purin type had a selective effect in the tests, and that on the other hand, the effect of the special inhibitors did not prove analogous with the results obtained in other tests known from the literature. Results obtained with synthetic cytokinins proved adequate, which confirms VAN OVERBEEK's assumption (1966) that the influence of plant hormones becomes effective through the induction of nucleic acid synthesis.

A comparative valuation of the effect of bioactive substances on the nitrogen metabolism suggests that the effect exerted on the protein level and protein synthesis could be further varied by the interrelation of the substances.

On the basis of the results obtained in the excitation tests we consider it justified to extend the nitrogen metabolism investigations to further fungus species representing more important taxonomic categories.

Studies on benzimidazole and its derivatives are thought to be important not only regarding mycelium development, but also taking the effect they exert on spore formation into consideration with special attention to the fact that some of the derivatives are systemic fungicides of high biological activity and special selectivity.

Research on plant hormone interrelations should be extended to include investigations into the complex nitrogen metabolism of host—parasite relations as well, for the primary purpose of making comparative evaluations concerning the extreme types of pathological resistance.

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METHOD OF CONTROLLING THE DEGREE OF PASTEURIZATION IN COW'S MILK- AND EWE-MILK PRODUCTS

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The author recommends the modified benzidine test — as well as the quick phosphatase test developed by Soviet experts and himself —, Storch's and Rothenfusser's peroxidase test and Lewerentz's test to control the degree of pasteurization in milk and milk products and points out that by the joint application of these methods a 5 minute heat treatment at 65°C as well as heating to 70°C, 75°C, 76°C, 78°C, 80°C and 90°C respectively can be controlled, and raw milk content demonstrated. The author emphasizes, further, the necessity of operative and technological controls besides taking the examination results in consideration, and the importance of the raw milk used as basic material being free of mastitis milk and of colostrum and cow's and ewe's milk in late lactation, sour and fermented milk in order to attain an unobjectionable pasteurization efficiency.

Introduction

On the basis of investigations carried out by BERKE (1933), BRIO *et al.* (1962) as well as at our own Institute, it became possible to establish the degree of pasteurization and to confirm the heat treatment not only of milk but also of the raw material of cow's milk- and ewe-milk products. Up to 1962 the special literature dealt with the control of pasteurization only in relation to milk.

It may be important to determine the pasteurized condition and the temperatures of pasteurization from 1. technological, 2. sanitary and veterinary, 3. nutrition biological, 4. commercial and 5. judical points of view.

1. From a technological point of view it is important because in the case of cheese-making a heat treatment of 72°C — or 63—65°C for 30 minutes —, in yoghurt-making of 90°C and in the case of cream pasteurization of 95°C is favourable.

2. Sanitary and veterinary aspects. According to our own investigations (1970) e.g. at a temperature of 90°C the "dry" and "wet" viruses of foot-and-mouth disease are killed in 35 seconds, while at 80°C in 70 seconds. If these data are at hand, and of the technical parameters of a pasteurizer the duration of temperature — assumed to be 70 seconds — is known, and a heating temperature of 90°C prescribed, with the method developed by LEWERENTZ (1964) we can control not only in milk but also in milk products whether they have

Table 1

Determination of percentage of heat-untreated matter and degree of pasteurization in various milk products (milk, cream, buttermilk, reconverted buttermilk powder, reconverted milk powder, butterplasm, ewe-milk)

Test / Milk product	Modified benzidine-test	Alkaline phosphatase test		Peroxidase-test		Lewerentz test	Evaluation
		time of incubation at 37°C		after Storch	after Rothenfusser		
		10–15′	2^h				
1 Milk, cream, buttermilk, reconverted buttermilk, reconverted milk, butterplasm, ewe-milk	non-transparent blue	carmine	carmine	blue	green	yellowish, yellowish-white	Milk, cream or ewe-milk raw, basic material heat-untreated
	transparent blue	pale pink	carmine	blue	green	yellowish, yellowish-white	5 per cent of the basic material untreated by heat
	transparent blue	pale pink	carmine	blue	green	yellowish, yellowish-white	1–2 per cent of the basic material untreated by heat
	transparent blue	white	pale pink	blue	green	yellowish, yellowish-white	Less than 1 per cent of the basic material untreated by heat
	transparent blue	white	white	blue	green	yellowish, yellowish-white	Heating temperature attained or exceeded 70°C, or was maintained for 5 minutes at 65°C
	of blueish tint	white	white	blue	green	yellowish,	Heating temperature attained 75°C
	of yellowish tint	white	white	blue	green	yellowish, yellowish-white	Heating temperature attained or exceeded 76°C
	of yellowish tint	white	white	blueish-grey, grey	greenish-brown	yellowish, yellowish-white	Heating temperature attained or exceeded 78°C

of yellowish tint	white	white	white	of yellowish tint	yellowish, yellowish-white	Heating temperature attained or exceeded 80°C
transparent blue	pale pink	carmine	blueish-grey, grey	greenish-brown	yellowish, yellowish-white	5 per cent of the basic material contained heat-untreated matter
transparent blue	pale pink	carmine	white	greenish-brown	yellowish, yellowish-white	1—2 per cent of the basic material contained heat-untreated matter
of yellowish tint	white	pale pink	white	of yellowish tint	yellowish, yellowish-white	Heat-untreated content of the basic material was below 1 per cent
of yellowish tint	white	white	white	of yellowish tint	rose-red	Heating temperature reached or exceeded 90°C. Milk powder was made of pasteurized milk or by cylinder drying, at 90°C
transparent blue	pale pink	carmine	blueish-grey, grey	greenish-brown	rose-red	Products pasteurized at 90°C or above contained 5 per cent non-heat-treated matter
transparent blue	pale pink	carmine	white	greenish-brown	rose-red	Products pasteurized at 90°C or above contained 1—2 per cent non-heat-treated matter
of yellowish tint	white	pale pink	white	of yellowish tint	rose-red	Products pasteurized at 90°C or above contained less than 1 per cent non-heat-treated matter

been heated at 90°C, which is important from the point of view of the prevention of epidemics and the checking of their spread.

3. In the case of favourable sanitary and veterinary conditions and when the technology does not require high temperatures of pasteurization, short time pasteurization carried out precisely and thoughtfully can be safely applied; its advantages from the points of view of nutritive value and taste were pointed out already by RIEVEL—FETTICK (1909).

4. It may be important to determine the degree of pasteurization in a commodity milk product due to the absence of medical and veterinary certificates, or those of quality and origin.

5. If temperatures prescribed by the authorities or under contract are not observed, during the legal procedure which follows expert opinion may be required.

Material and Method

Models were prepared for the examinations under laboratory conditions with an operative technology, by mixing raw milk with pasteurized milk samples at various ratios; these mixtures and the products made from them were examined. To determine the degree of pasteurization the following investigation methods were used:

1. Benzidine test modified at our Institute. It differs from the test generally known from the literature in employing Rothenfusser's lead acetate serum (cit. WINKLER *et al.* 1930) instead of milk for the examination, by which a more intensive and readily evaluated colour reaction can be obtained.

2. Quick and simplified alkaline phosphatase test developed by the above mentioned Soviet experts and by the author (WAGNER 1965).

3. Storch's and Rothenfusser's peroxidase test (WINKLER *et al.* 1930).

4. Lewerentz's test (LEWERENTZ 1964), the application of which was extended to study cream and other milk products too (WAGNER 1968).

Results

The results are contained in Tables 1, 2, 3, 4, 5 and 6. The tables reveal that no method applied by itself gives reliable results. No safe information can be obtained on the heat treatments in this way. Reliable results are only given by the parallel application of various methods, especially when the origin of the sample is unknown. Methods which give preference to the technological and operative control and consider laboratory examinations performed on the spot as indispensable complements are more reliable even if temperature and its maintenance are controlled automatically, since e.g. in plate pasteurizers a hair-crack, or a technical defect may result in raw milk getting into the pasteurized milk through the heat exchanger even with the prescribed temperature maintained.

With the suggested methods we can find out whether cow's milk and -products and ewe milk and -products have been kept for 5 minutes at 65°C or heated to 70°C, 75°C, 76°C, 78°C 80°C, and 90°C.

Table 2

Percentage of non-heat-treated matter and degree of pasteurization in various milk products (sour cream)

Test / Product	Modified benzidine-test	Alkaline phosphatase-test		Peroxidase-test		Lewerentz test	Evaluation
		time of incubation at 37°C					
		10 – 15′	2^h	after Storch	after Rothenfusser		
2 Sour cream	—	carmine	carmine	grey	—	yellowish, yellowish-white	Sour cream was made of non-heat-treated basic material
	—	pale pink	carmine	grey	—	yellowish, yellowish-white	Sour cream contained 5 per cent non-heat-treated matter
	—	white	pale pink	grey	—	yellowish, yellowish-white	Sour cream contained less than 5 per cent non-heat-treated matter
	—	white	white	grey	—	yellowish, yellowish-white	Heat treatment attained or exceeded 70°C
	—	white	white	white	—	yellowish, yellowish-white	Heat treatment attained or exceeded 80°C
	—	pale pink	carmine	white	—	yellowish, yellowish-white	Products pasteurized at 80°C or above may contain 5 per cent non-heat-treated matter
	—	white	pale pink	white	—	yellowish, yellowish-white	Products pasteurized at 80°C or above contained less than 5 per cent non-heat-treated matter
	—	white	white	white	—	rose-red	Heat treatment attained or exceeded 90°C
	—	pale pink	carmine	white	—	rose-red	Products pasteurized at 90°C or above contained 5 per cent non-heat-treated matter
	—	white	pale pink	white	—	rose-red	Products pasteurized at 90°C or above contained less than 5 per cent non-heat-treated matter

Table 3

Percentage of non-heat-treated matter and degree of pasteurization in various milk products (yoghurt, kefir)

Test / Product	Modified benzidine-test	Alkaline phosphatase-test		Peroxidase-test		Lewerentz test	Evaluation
		time of incubation at 37°C		after Storch	after Rothenfusser		
		10–15′	2^h				
3 Yoghurt, kefir	—	pale pink	pale pink	grey	—	yellowish, yellowish-white	The product was made of non-heat-treated basic material
	—	white	white	grey	—	yellowish, yellowish-white	Heat treatment attained or exceeded 70°C
	—	white	pale pink	grey	—	yellowish, yellowish-white	The product contained 5 per cent non-heat-treated matter
	—	white	white	white	—	yellowish, yellowish-white	Heat treatment attained or exceeded 80°C
	—	white	pale pink	white	—	yellowish, yellowish-white	The product contained 5 per cent non-heat-treated matter
	—	white	white	white	—	rose-red	Heat treatment attained or exceeded 90°C
	—	white	pale pink	white	—	rose-red	Products pasteurized at 90°C contained 5 per cent non-heat-treaed matter

Table 4

Percentage of non-heat-treated matter and degree of pasteurization in various milk products (whey, reconverted whey powder, sour buttermilk, reconverted sour buttermilk)

Test / Product	Modified benzidine-test	Alkaline phosphatase-test		Peroxidase-test		Lewerentz test	Evaluation
		time of incubation at 37°C		after Storch	after Rothenfusser		
		10–15′	2^h				
4 Whey, reconverted whey powder, sour buttermilk, reconverted sour buttermilk powder	—	pale pink	pale pink	blue	green	yellowish, yellowish-white	The product was made of non-heat-treated basic material
	—	white	white	blue	green	yellowish, yellowish-white	Heat treatment attained or exceeded 70°C
	—	white	white	grey	greenish-brown	yellowish, yellowish-white	Heat treatment attained or exceeded 78°C, or the products contained 2 per cent non-heat-treated matter
	—	white	white	white	white	yellowish, yellowish-white	Heat treatment attained or exceeded 80°C
	—	white	white	white	white	rose-red, purplish	Heat treatment attained or exceeded 90°C
	—	white	white	grey	greenish-brown	rose-red, purplish	Products treated at 90°C contained 2 per cent non-heat-treated matter

Table 5

Percentage of non-heat-treated matter and degree of pasteurization in various milk products (quarg, curded ewe-cheese)

Test / Product	Modified benzidine-test	Alkaline phosphatase-test		Peroxidase-test		Lewerentz test	Evaluation
		time of incubation at 37°C					
		10–15′	2^h	after Storch	after Rothenfusser		
5 Quarg, Curded ewe-cheese	—	pale pink	pale pink	grey	—	yellowish, yellowish-white	**The product was made of non-heat-treated basic material**
	—	white	white	grey	—	yellowish, yellowish-white	**Heat treatment attained or exceeded 70°C**
	—	white	white	white	—	yellowish, yellowish-white	**Heat treatment attained or exceeded 80°C**
	—	white	white	white	—	rose-red	**Heat treatment attained at least 90°C**

With an efficient pasteurization in view not only the prescribed temperatures have to be observed. Milk must be free of mastitis milk as well as of colostrum and cow's and ewe's milk in late lactation or sour components as they reduce the efficiency of pasteurization (Rievel—Fettick 1909, Nyiredy-Rudnyánszky 1967).

Table 6

Percentage of non-heat-treated matter and degree of pasteurization in various milk products (Gruyere cheese, processed cheese)

Test Product	Lewerentz test	Evaluation
6 Gruyere and processed cheese	yellowish, yellowish-white	Processing temperature did not exceed 90°C; post-heating at 56°C following pasteurization at 72°C did not take more than 45 minutes
	rose-red	Processing temperature exceeded 90°C; post-heating at 56°C following pasteurization at 72°C took more than 45 minutes

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IMPORTANCE OF MALE CONTROL IN PREVENTING DAMAGE DONE BY THE SAN JOSÉ SCALE (QUADRASPIDIOTUS PERNICIOSUS COMST.)

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Under the scute, during pupation the males of the San José scale are sensitive to certain plant protectives, thus to organic phosphorus preparations tested at our Institute as well. During the development of the overwintering generation — unlike conditions existing in the summer generations — at a given time the majority of the individuals within the population is at the same stage of development, males get into a state of sensitivity to plant protectives within a relatively short period. By intervening before the beginning of the imagos' swarming, the majority of the males can be destroyed. As a result of this — since San José scales do not multiply by parthenogenesis — the larvae can be prevented from appearing in masses. At present the procedure is of importance first of all in those stone fruits in which the appearance of the larvae of the overwintered generation coincides with the ripening time of the fruits.

Introduction

With the present possibilities for chemical control an intensive application of insecticides is required for the prevention of damages done by the San José scale. Infection-free fruits can be produced only in orchards where regular control can be ensured from the appearance of the larvae of the overwintered generation till the end of the summer and by this the larvae of the summer generation can also be prevented from establishing themselves. However, owing to the ripening time of fruits such a high degree of control can only be ensured in winter apple and winter pear plantations. In the case of earlier fruit species and varieties a decreased efficiency of control has to be reckoned with. Chemical control has a minimum possibility in cases when the ripening of fruits and appearance of larvae from the overwintered generation occur at the same, or nearly the same time.

Specific control operations against various pests, such as e.g. the sterile male technique, will be difficult to carry out in those orchards where within the same vegetation period insecticides must be applied against the San José scale on more than one occasion.

To promote the solution of the existing problems we began to study new possibilities of protection against the San José scale, e.g. the control of males.

NEWCOMER—YOTHERS (1929), YOTHERS (1940) were the first to point out that the male San José scales were sensitive to calcium sulphite even under the scute, and after their destruction the unimpregnated females would die later without progenies.

At the International Scientific and Technical Conference held in 1959 in Prague on the problems of Plant Protection and Quarantine Service it was suggested that as a complementary procedure against San José scale, spraying against males — according to the idea of that time with chlorinated hydrocarbons — would be worth being included in the spraying programme of fruits (ANONYM 1959).

During our work we studied the effect of various preparations — first of all of those containing organic phosphorus — on the different development stages of males, as well as the optimum time of their application.

Material and Method

The phenology of males was studied on bark samples collected in 1962 from apple trees (Zalaapáti) while in 1965 and 1966 from sour cherry trees (Tárnok), from the middle of March or beginning of April till November once a week, while during the development of the overwintering generation every three days from the expected time of pre-pupae development.

In the control experiments a shoot in each of four trees was treated at a length of 50—60 cm with each preparation examined. In 1969 3 adjacent rows were treated with an S-293-type sprayer furnished with a hand sprinkler in a sour cherry plantation of about 0.2 ha with medium high trunks. In 1970, in a spindle bush plantation (sour cherry) treatments were applied at the infected points of the plot by a motor sprayer supplied with automatic ventilator (Holder). In the course of a methylparathion spraying, which became necessary four days later against other pests in the same plantation, the previously treated surfaces could not be omitted for technical reasons.

Spraying was carried out in May, immediately before the swarming of the males, when they were already in the stage of prepupation or pupation. On one occasion in 1967 the effect of spraying carried out in July was also studied.

Investigations were carried out with plant protectives belonging to the following active agent groups: Formothion (Anthio), Tiometon (Ekatin), nicotin, 00,-Dimethyl-S-(2-metoxy-1,3,4-thiadizol-5 (4H)-onyl-(4)-methyl)-dithio-phosphate (Ultracid). Treatments in 1969 and 1970 were carried out with Ultracid 40 applied.

Effects exerted by the insecticides on males were studied five days after spraying, when the ratio of destroyed to living pupae was determined with 25 individuals per replication. Later, at the time of the mass appearance of the larvae the ratio of impregnated females to unimpregnated ones (females with embryo and without) was evaluated.

Results

According to the data of the phenological survey, under conditions in Hungary the different development stages of the overwintering generation showed the following trends depending on the weather conditions. Between April 22nd and May 15th pre-pupae occurred for not less than 14 and not more than 18 days; between April 26th and May 19th pupae for not less than 15 and not more than 18 days. The earliest day when males began to fly was April

30th and the latest May 9th; flying lasted for about 14—19 days. During the development of the summer generation male pre-pupae, pupae and imagos of different ratios and quantities occurred constantly till the end of the vegetation period (Sheta—Jenser 1970), (Fig. 1).

Of the insecticides tested in the course of the experiments Ultracid proved to be the most effective. A somewhat lower percentage was destroyed by Ekatox 20, while Tinox and Anthio as well as nicotine were of a poorer effect. As to stage-specific effects on pre-pupae and pupae no significant differences were found, nevertheless it is worth mentioning that the preparations tested — with the exception of Tinox — exerted the highest toxic effect on the pupae (Tables 1 and 2).

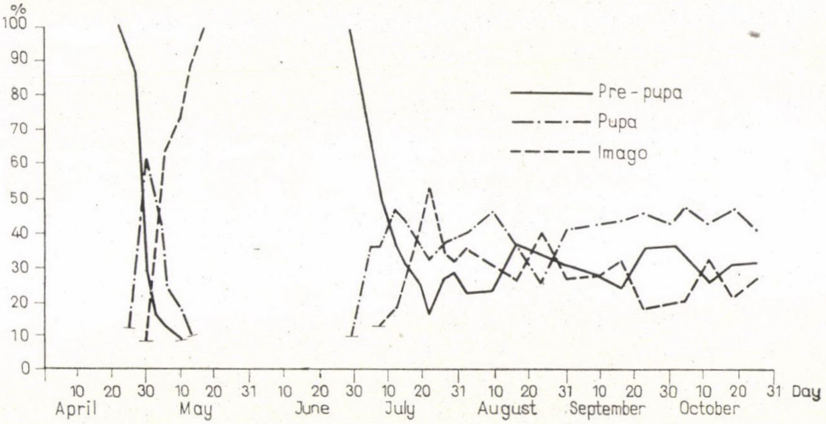

Fig. 1. Proportions of development stages of male San José scales (Tárnok, 1966)

Table 1

Stage-specific effects of certain plant protectives on male San José scales at the stages of pre-pupa and pupa
Tárnok, 1967

Preparation	Pre-pupa		Pupa	
	5 May	4 July	5 May	4 July
Anthio 0.1%	66.0 ± 1.35	—	71.55 ± 1.1	
Nicotin 0.1%	—	55.6 ± 1.41	—	65.58 ± 1.0
Parathion 20 0.2%	—	61.0 ± 1.73	—	69.90 ± 1.3
Tinox 0.1%	70.3 ± 1.25	—	68.89 ± 1.2	—
Ultracid (Supracid) 0.1%	71.5 ± 1.49	67.1 ± 1.55	77.35 ± 1.3	75.96 ± 1.9
Control	10.2 ± 1.07	7.7 ± 1.01	9.48 ± 1.1	11.34 ± 1.8

The proportion of females unimpregnated following the treatment depended on the degree of infection in the neighbouring trees and on the location of the treated trees. When the experimental spraying was applied to a relatively small surface of the trees, to certain parts of the shoots, the activity of males coming from untreated surfaces also had to be reckoned with. A similar effect was observed in cases when spraying was applied to larger surfaces. On trees in the inner rows of the treated plot, where the extent of males flying in could not be considerable, the proportion of impregnated females decreased to a minimum. On treated trees either in the marginal row or in an infected environment the proportion of impregnated females amounted to 52-56 per cent, on untreated trees practically all females became impregnated (Table 3). After spraying applied to a continuous surface in a spindle bush orchard the San José scale infection was practically terminated.

Table 2

Proportion of unproductive females of the San José scale after treatments carried out at the swarming time of males with various plant protectives
Tárnok, 1966

Preparation	Total number of females	Productive	Unproductive	
	number	number	number	percentage
Anthio 0.1%	384	103	281	73.33±1.3
Tinox 0.1%	371	73	298	80.37±1.09
Ultracid (Supracid) 0.1%	424	56	368	86.66±1.37
Control	369	320	49	8.01±1.19

Table 3

Effect of Ultracid application against male San José scales on the proportion of productive females
Tárnok, 1969

Place and number of trees evaluated	San José scales evaluated total number	Females with embryo percentage
Inner row of the plot 8 trees	633	3.05± 3.8
Marginal row 3 trees	147	52.80±15.3
Treated tree in infected environment 1 tree	44	56.8
Untreated 3 trees	500	97.7 ± 2 3

Discussion

Males of the San José scale proved sensitive under the scute to calcium sulphite (Yothers 1940), chlorinated hydrocarbons (Anonym 1959) and — according to our investigations — certain insecticides containing organic phosphorus. According to the unanimous results of some authors (Yothers 1940; Müller 1952; Gentille—Summer 1958) and our own investigations San José scales do not multiply through parthenogenesis. Unimpregnated females, though remaining alive for a considerable time, die without progenies. Thus, with protection against males population density can be reduced to a minimum and damages prevented. The possibilities of applying this method are to some extent limited as yet, due partly to the way of action of the male-toxic insecticides tested so far, partly to the phenology of San José scale and plants to be protected.

With Hungarian conditions taken in consideration, the opportunity for an efficient intervention arises when the males of the overwintering generation develop. Namely, the development of the first individuals overwintering as larvae is continued almost at the same time in spring, so at the end of April or beginning of May the males enter the stages of pre-pupa, pupa and imago with relatively small differences in time. Treatments applied immediately before the development of the imagos destroy the majority of the males. During the development of the summer generation, from the beginning or middle of July till the autumn frosts all developmental forms of the San José scale occur, therefore population density can be influenced only by repeated interventions at the most.

The control of males in the protection of apple is not possible for the time being. The stages of pre-pupa, pupa and imago of males in the overwintering generation coincide with flowering in apple, and the preparations so far tested and found effective are dangerous to bees. Between the end of flowering in pear and the beginning of swarming of the males there is an interval of 1—2, sometimes even 10—12 days. Between flowering in apricot, peach and plum and the development of males there is also sufficient time to carry out control operations.

The utilization of the possibility of protection against males is considered especially important in the case of cherry and sour cherry, because the ripening of fruits in some of their varieties coincides with the mass appearance of San José scale larvae. Treatment of early varieties is not possible on account of the long waiting time of the otherwise efficient insecticides. According to the data available there is an interval of 2—4, sometimes even 12—14 days between the end of flowering in cherry and sour cherry and the beginning of swarming of males in most years.

For the purpose of control those preparations are considered to be primarily suitable which — while able to destroy living pre-pupae and pupae under the scutes — have a contact effect too and are also toxic to swarming males. In our opinion, this is also the explanation for the high degree of protection provided by Ultracid.

The above data give sufficient evidence of the fact that a great possibility for protection against San José scale is provided by the control of males in the overwintering generation, by which the number of later treatments with insecticides can be reduced. Further investigations are required to find preparations by the use of which the control method in question can be extended to other plants too, and a prevention of the damages done by the San José scale ensured with the least possible disturbance of biocoenosis.

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MICROELEMENT CONTENT IN LUCERNE HAYS

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The crude nutrient — as well as the major macro- and microelement contents per cutting were examined in lucerne hays obtained from mantle-sand- and czernozem type soils. The data showed that lucerne hay produced in the mantle-sand soil had in each cutting a lower pure protein- and higher row fiber content than that produced in czernozem soil. Irrespective of the soil type, hays of three-year-old lucernes contained less Ca, K and Na than those of two-year-old ones. No significant difference in the microelement content of three-year-old plants was found between the two soil types. The Mn and Zn contents in the hay of two-year-old lucerne plants were, however, lower. The ratio of Fe : Mn : Zn : Cu : Mo = 80 : 40 : 20 : 5 : 1 considered suitable from the point of view of feeding was not attained in the lucerne hays examined due to their low Mn and Zn contents, thus they were of no full value biologically implying that a supplementary application of Mn and Zn may be justified.

Introduction

Lucerne hay, the most important Hungarian rough fodder is a significant protein- and vitamin A-source for livestock. Owing to its role played in feeding it is important to know — beside its carotene content — also the mineral substances and microelements contained in it.

The microelement content in plants is equally influenced by the soil type, the climatic and precipitation conditions as well as the fertilization system employed in the respective area. The examination of the mineral and microelement contents of lucerne hays produced in the two dominant soil types of the region was, therefore, considered to be of interest. Since lucerne is a perennial plant and — depending on the cultural practices applied — is cut more than once a year, the investigations described here were made with samples obtained from the first three cuttings of two- and three-year-old lucernes generally utilized in practice.

Data on the microelement content of hays can be found in the Hungarian literature in publications by Tölgyesi (1965), Baumann (1950), Mócsy—Tölgyesi (1959, 1960) and Haraszti (1966a, 1966b). The microelement content in plants grown in alkali soils was studied by Módor—Tölgyesi (1964, 1965). Microelement problems related to the flora of moors were dealt with by Belák *et al.* (1969, 1970) and Szalai *et al.* (1970a, 1970b). The macro- and microelement uptake of plants grown in acidic and calciferous sand soils was studied by Tölgyesi *et al.* (1970). Trends of the macro- and microelement contents of lucernes grown in various soil types were first discussed in Hungary by Szentmihályi (1963). Papers published by Mócsy—Tölgyesi (1959, 1960) and Tölgyesi (1965) provided data on the microelement content of lucerne on the basis of a great number of analysed samples originating from various parts of the country. The authors found an average of 44.1 (12.5—101) ppm Mn content, 9.8 (4.2—15.2) ppm Cu and 0.13 (0.07—0.26) ppm Co content. As to seasonal changes in the microelement contents of lucernes they found that lucerne had a 7—8 per cent higher microelement content when cut in May than when cut later.

Material and Method

For the analyses samples were collected from lucerne stands grown under dry conditions in plots with various soil types in the Borbáspuszta and "Parasztfőiskola" farm units of the Institute's farm. The important meteorological data of the vegetation period in the experimental area are shown in Table 1. Thus, two- and three-year-old plants from lucerne plots of lime-covered czernozem soil type as well as three-year-old lucerne plants grown in a mantle-sand soil type were analysed. The basic examination results of the two soil types are contained in Table 2. The samples were taken from plants windrowed on each occasion in the following way. From handfuls of lucerne plants collected at random at ten different places of the plot a sample sheaf was formed. Five sample sheaves per plot were collected in each cutting.

The sample sheaves were dried first at air temperature then at 60°C, and ground. The analyses were made with air-dried ground plants. The crude nutrients were determined on the basis of feedstuff standards, while the macro- and microelements by the ashes of the plants: Ca, K and Na with flame-photometry, Mg with colorimetry. Fe, Mn, Zn and Cu were studied with a polarograph (Prohászka—Horváth 1970, MNOSZ 1953). Mo was determined in the complex form of molybdic rhodamid, reduced with $SnCl_2$ and extracted with a mixture of ether and toluol (Tölgyesi 1969).

Results

The crude nutrient content per cutting of lucerne hay produced in the two soil types is presented in Table 3. The data of the table show that differences worth mentioning between the plants of the growing site in question are only found in pure protein- and raw fiber contents. Lucerne hay produced in a mantle-sand soil had in each cutting a lower pure protein and higher raw fiber content than that produced in a czernozem soil. The higher pure protein content of the latter hays is combined with a lower amid content, which suggests that they are biologically more valuable feedstuffs.

The trends shown by the macro- and microelement contents of lucerne hays can be seen in Table 4. When studying the table we find that — irrespective of the soil type — the three-year-old lucernes contained less Ca, K and Na than the two-year-old ones. The Ca and Na contents of the former hays increased with cutting carried out later. In the case of K a decrease was observed.

Table 1

Precipitation and air temperature of the experimental area in the vegetation period on the basis of data obtained from the Agrometeorological Observatory at Kecskemét

Designation	Months											
	I.	II.	III.	IV.	V.	VI.	VII.	VIII.	IX.	X.	XI.	XII.
mm precipitation on a sixty years average (1901—1960)	28.0	31.0	33.0	47.0	59.0	60.0	53.0	45.0	47.0	47.0	52.0	40.0
mm precipitation in the year of the study (1969)	19.1	104.2	18.6	24.9	15.7	169.1	28.8	82.1	8.9	17.2	58.1	112.6
Deviation from the mean	−8.9	+73.2	−14.4	−22.1	−43.3	+109.1	−14.2	+37.1	−38.1	−29.8	+6.9	+72.6
C° mean temperature on a sixty years average (1901—1960)	−1.6	−0.1	5.5	11.1	16.7	20.0	22.1	21.1	16.7	10.8	4.9	0.8
C° mean temperature in the year of the study (1969)	−3.2	−0.5	3.8	10.8	19.1	18.6	21.4	19.8	17.0	11.6	8.9	−2.4
Deviation from the mean	+1.6	+0.4	−1.7	−0.3	+2.4	−1.4	−0.7	−1.3	+0.3	+0.8	+4.0	+1.6

Table 2

Basic analysis data of mantle-sand and czernozem soils

Soil type	Depth of sample cm	hy	$CaCO_3$ %	pH		Humus %	Available	
				H_2O	KCl		P_2O_5	K_2O
							mg/100 g soil	
Mantle-sand	0— 20	0.95	2.0	7.1	6.9	0.82	7.6	5.9
	20— 30	0.80	3.0	7.0	6.8	0.75	—	—
	30— 60	1.60	5.5	7.5	7.0	1.60	—	—
	60—150	1.10	22.6	8.2	7.7	0.50	—	—
Czernozem soil	0— 20	2.70	10.0	7.7	7.4	2.35	15.2	12.5
	20— 40	1.8	18.2	8.2	7.8	1.50	—	—
	40—150	1.2	20.5	8.5	8.2	0.60	—	—

Note: — = not analysed

Table 3

Trends of crude nutrient contents in lucerne hay per cutting, in different soil types

Time of sample taking	Crude nutrients %	Origin of samples		
		Mantle-sand	Czernozem	
		3-year-old lucerne stand	3-year-old lucerne	2-year-stands
	Crude protein	21.25	20.50	20.31
	Pure protein	13.50	14.60	13.30
First cutting	Raw fiber	28.40	27.30	24.50
	Ash	8.30	8.80	9.60
	Amides	7.75	5.90	7.01
	Crude protein	21.87	24.37	24.37
	Pure protein	11.60	12.50	16.50
Second cutting	Raw fiber	25.20	28.50	26.90
	Ash	7.90	7.60	8.50
	Amides	10.27	11.87	7.87
	Crude protein	20.62	20.62	20.50
	Pure protein	10.80	13.80	14.20
Third cutting	Raw fiber	26.00	23.60	25.00
	Ash	8.10	7.80	7.80
	Amides	10.82	6.82	6.30

The Mg content gradually decreased in lucernes grown in mantle-sand soil, while increased up to the third cutting in those grown in czernozem soil. In two-year-old lucernes the Ca and K contents decreased while the Mg and Na increased in subsequent cuttings.

No essential difference in microelement content was found between the three-year-old plants of the two soil types, but the Mn and Zn contents of the two-year-old lucerne hays were lower. Changes observed during the vegetation period were also much more unambiguous. E.g. Mn and Zn considerably decreased in subsequent cuttings with both soil types and lucerne hays.

Table 4

Trends of macro- and microelement contents in lucerne hays per cutting, in different soil types

Origin of samples	Time of sample taking	Ca	Mg	K	Na	Fe	Mn	Zn	Cu	Mo
		(...... g/kg ......)				(...... mg/kg ......)				
3-year-old lucerne stand in mantle-sand soil	First cutting	12.4	2.48	10.2	3.4	402	30.0	22.9	10.1	1.25
	Second cutting	11.1	2.35	17.9	3.2	340	23.2	21.0	9.0	1.72
	Third cutting	17.0	2.19	8.8	3.9	408	22.1	16.0	10.7	2.68
3-year-old lucerne stand in czernozem soil	First cutting	13.4	3.25	11.9	3.5	417	30.0	32.5	9.0	1.96
	Second cutting	11.1	3.11	9.5	3.9	435	25.8	20.4	9.0	2.08
	Third cutting	15.3	4.67	5.0	5.7	380	20.9	20.1	10.2	1.70
2-year-old lucerne stand in czernozem soil	First cutting	17.4	2.25	14.6	4.6	375	25.6	21.0	10.1	3.32
	Second cutting	14.2	2.17	13.4	4.8	402	23.3	18.0	11.0	2.94
	Third cutting	13.2	3.04	7.7	5.8	452	17.5	16.0	11.4	2.72

Lucernes grown in a mantle-sand soil had a constant Fe content during the subsequent cuttings, while in a czernozem soil Fe considerably decreased in three-year-old and increased in two-year-old plants.

The Mo content of lucerne hays only showed an increase in the mantle-sand soil, in czernozem it decreased; Cu showed the least change in both soil types during the subsequent cuttings. In the Hungarian literature a similar statement — namely, that the microelement content of lucerne cut in May is higher than of those cut later — can be found (Mócsy—Tölgyesi 1959, 1960, Tölgyesi 1965).

Apart from the quantities of microelements their ratio is also important from the point of view of feeding. In relation to the dry-matter content of feedstuffs the following microelement ratios were recommended by Tölgyesi: Fe : Mn : Zn : Cu : Mo = 80 : 40 : 20 : 5 : 1. According to our analyses trends shown by these elements per cutting in the two soil types were as presented in Table 5. It is seen that lucerne hay obtained from any one of the cuttings cannot be considered as of full biological value, and the ratio of elements becomes worse in each subsequent cutting.

Due to its low Mn content lucerne hay is objectionable in most soil types from a feeding point of view. In the present case even a Zn deficiency is added to it. In the case of two-year-old lucernes a Cu deficiency also seemed to occur, since the Cu/Mo ratio of 5 or over decreased to a ratio of 3 or 4. The Cu deficiency in question is, however, only secondary, as the 9—11 ppm Cu content of hays obtained from two-year-old lucerne plants is considered satisfactory, and the low ratio of Cu/Mo is only due to the high Mo content.

According to the data of our analyses in the case of the above lucerne hays a supplementary application of Mn and Zn may be justified.

Table 5

Trend of microelement ratios per cutting in different soil types

Origin of sample	Time of sample taking	Fe	Mn	Zn	Cu	Mo
3-year-old lucerne in mantle-sand	First cutting	319	23	18	8	1
	Second cutting	197	13	12	5	1
	Third cutting	152	8	6	4	1
3-year-old lucerne in czernozem soil	First cutting	212	15	16	5	1
	Second cutting	209	12	10	4	1
	Third cutting	223	12	12	6	1
2-year-old lucerne in czernozem soil	First cutting	113	8	6	3	1
	Second cutting	137	8	6	4	1
	Third cutting	166	6	6	4	1

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THE DETERMINATION OF THE DIGESTABILITY OF SOME FEEDSTUFFS IN CHICKENS USING CHEMICAL METHOD

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Eight digestion trials with triplicate cocks have been undertaken to determine the feeding value of feedstuffs used in the feeding of poultry. Five materials have been done alone, with other three (cotton seed meal, wheat bran and lucerne dried) the maize was used as a basal ration. For determining the urinary nitrogen of the mixed excrement the chemical method has been applied. The composition, the digestion coefficient and the feeding value of these feedstuffs are discussed here as well as its relations to similar foodstuffs.

Introduction

The fact that the chicken excertes urine and faeces together has greatly complicated digestibility studies. Considerable difficulties arise with the various foodstuffs in this respect. In most of works done on digestibility in the chicken the methods are not fully dependable for protein has been used and some of the data so obtained are rather different. Therefore it is apparent that some means must be devised for separating the faecal and urinary nitrogen in poultry excreta before the digestibility of nitrogen can be determined. Research in avian physiology has been hampered by the lack of satisfactory method for separation of urine and faeces in birds. A few investigators have used birds on which the faecal and urinary outlets were separated by surgical means and for this purpose a number of surgical methods has been described. The most popular method was the anus praeternaturalis, but this was limited largely to mature birds. Voltz (1909) used the artificial anus but it is the work of Katayama (1924) that was most widely cited. He operated two cocks and determined the digestibility of various foodstuffs by direct and chemical method. Stotz (1931) also developed a chemical method for determining urinary nitrogen of mixed excrement and made use of the artificial anus to check the method. He gave the highest recovery and this recovery amounts only to 86–95 per cent of the urine – free faecal nitrogen. On the other hand, he found that during the nitrogen determination according to his procedure uric acid was completely removed from the sample and it seemed probable that the protein digestion coefficient was generally somewhat too high.

Daikow (1932) described a method for collecting urine from a hen without surgery. He dissolved all the uric acid in ammonia from mixed excrement and determined the nitrogen left in the residue, which he assumed to be undigested protein. **Fraps** (1944) determined total uric acid and ammonia nitrogen, substracted the sum from the total nitrogen and assumed the difference to be undigested nitrogen. **Mueller** *et al.* (1956) used the method developed by **Stotz** to determine the effect of age and sex on digestibility. They found the greatest differences between digestion coefficients for fibre and protein. In that connection **Fraps—Carlyle** (1939) reported that chicks fed ad-libitum a diet containing approximately 19 per cent of protein retained 42.5 per cent of the protein consumed during the period of approximately one to 4 weeks of age, 38.1 per cent during the period from one to 7 weeks of age and 33.1 per cent during the period from one to 13 weeks of age. **Osm** *et al.* (1957) used the direct method for the determination of digestibility in growing chickens fed purified and practical type diets.

On the other hand, examinations of the digestibility of other chemical constituents showed that sugars, dextrins and starch were completely digested but cellulose and lignine were indigestible. Crude fibre digestion for many different feeds was reported by **Kaupp—Ivey** (1922). **Fraps** (1928), **Titus** (1939), **Halnan** (1928) and **Mangold** (1934) presented reviews on the role of fibre in poultry feeding. **Henery** *et al.* (1955) found that the digestion of crude fibre by turkeys was negligible. Recently, **Titus** (1961) has showed that when the bulkiness of foodstuffs is compared with their fibre content, no marked relationship can be found between them and he mentioned 3 to 5 per cent crude fibre as sufficient for physical properties.

In an experiment on the digestion of different levels of fat Barred Plymouth Rocks breed, **Whitson** *et al.* (1934) found a significant increase of fat absorption from 8 to 10 and 12 weeks of age, when diets containing 3 to 9 per cent of fat were fed but not when diet contained 20 per cent fat. **Bolton** (1963) mentions that chicks digest and tolerate high levels of fat.

These differences make questionable any direct application to poultry of the results of digestion experiments obtained in cattle, sheep and swine of adult age. For this reason this work is undertaken to give reliable data on the digestibility of common foodstuffs used in poultry rations in Hungary.

Material and Method

Eight digestion trials were carried out in the Service of the Faculty of Agriculture, Keszthely, Hungary. Using three cocks in each of Rhode Island breed, the cocks were placed in individual cages in an "only" hen battery in such a manner that each cock could eat only the feed provided in its individual section.

The birds were given known amount of feed each day based on the daily average amount consumed during the preceeding 5 days. There was no refused or scattered food during the collection period. The maize was used as a basal ration with three foodstuff materials (cotton seed meal, wheat bran and lucerne dried), but the other feedstuffs (maize, wheat, barley, soya bean meal and peanut meal) were given alone. Materials were of commercial origin commonly fed in Hungarian livestocks.

During the experiment the animals were in health and good condition, they were weighed at the end of the experiment to make sure that they were maintaining their weight.

Droppings were collected daily on polyethylene film weighed and dried in a heated oven at 65—70°C for 16 hours and at 105°C for 3 hours, then weighed and finely ground in a mill. The 5 daily collections were combined to form a composite sample for each bird.

The analysis of foodstuffs and faecal material followed the conventional methods (1 and 12) using duplicate samples of 2—3 gm. each. Moisture was determined at 105°C.

Ekman *et al.* (1931) and Hartfiel (1961) procedures are used to get faecal protein content by using uranyl acetate $[UO_2(CH_3COO)_2 \cdot 2H_2O]$.

For calculating the organic substance (dry matter—crude ash) of the urine-free faeces the following formula was used: organic substance of faeces = organic substance of excrete—organic substance of urine, where organic substance or urine = 3.0 nitrogen of urine according to Daikod (1932). The fat content of the urine free faeces was calculated by the following formula: Fat of faeces = fat of excreta — fat of urine where fat of urine = 0.032 × organic substance of urine according to Engler (1933).

The methods to calculate the digestion coefficient and starch value based by Kellner (1926) and followed by Ghoneim *et al.* (in press), the methods for the calculation of total digestible nutrients used by Maynard—Loosli (1951) and followed by Darwish *et al.* (in press) were applied.

Results

1. *Maize.* The composition of the maize indicates that it contains a high amount of N. F. E. (Nitrogen Free Extracts), 76.08 per cent, a moderate content of crude protein (11.30 per cent), a low amount of crude fibre (2.44 per cent) and crude fat (4.64 per cent), similarly to that published by Titus (1961), Bolton (1963) and similar to the Hungarian Standars (MSz 6830T/65 I. v.) but it differs in crude fibre content from that obtained by Ghoneim (1958).

The digestion coefficients of its nutrients (excluding crude fibre) are of a high order being higher than 82 per cent, in case of crude fibre it is too low (25.56 per cent) being higher than the data of Titus (1961).

The feeding value of the maize is 71.65 Starch Value (S. V.) and 73.21 Total Digestible Nutrients (T.D.N.) as fed being slightly lower than the S.V. calculated from the data of Titus (1961) which is 78 S.V. (80 per cent T.D.N.). The Metabolizable Energy (M.E. of the maize is 1200 Kcal/lb (533 Kcal/kg), being lower than the value obtained by Bolton (1963) which was 1500 Kcal/lb (671 Kcal/kg).

On the other hand, the feeding value resulted here is too low when compared with that obtained by Ghoneim (1958) who — instead of cock — used sheep in performing the digestible trial, therefore his value was 82.03 S. V. as fed (89. 18 T.D.N).

The digestible protein content has proved to be 8.27 per cent. The maize is widely used in the feeding of poultry. It is consumed with preference and readily digestible, it has a low content of crude fibre and supplies a larger quantity of total digestible nutrients than most other foodstuffs commonly used in poultry feeding.

2. *Wheat.* The composition of the wheat indicates that the major nutrient is the N.F.E. (81.18 per cent) and there is medium content of crude protein (11.73 per cent). It contains also a low amount of fat (2.41 per cent) and crude fibre (1.95 per cent). The composition is similar to that published by the previous workers (Bolton 1963, Ghoneim 1958, MSz. 6830T/65 I. v., Titus 1961).

The digestion coefficient for N.F.E. and crude protein characteristic of its feeding value, is of high order being 84.89 per cent and 84.64 per cent, respectively. It is about 70 per cent for crude fat and lower for crude fibre, about 19 per cent but higher than the value of Titus (1961): 9 per cent.

The feeding value was found 70.38 S. V. as fed (71.51 T.D.N.), similar to that recorded by Titus (1961), which were 72.04 S. V. and 73.0 T.D.N. The metabolizable energy of wheat is 1176 Kcal/lb (523 Kcal/kg), slightly lower than the value of Bolton (1963), which is 1380 Kcal/lb (613 Kcal/kg).

There is an agreement between our starch value and that obtained when sheep were to perform digestibility trial by Ghoneim (1958). It contains 8.57 per cent digestible protein as fed.

Generally, large quantities of wheat have been used in poultry feeding. It is very preferable and is an excellent foodstuff. It supplies the same total digestible nutrients and digestible protein as maize and usually is a somewhat better source of most of the vitamins of the B-Complex.

Too high content of gluten is not desirable in feeding wheats, for the crop content tends to become doughy, leading to digestive disorders. Newly harvested wheat has harmful effect on flows, which appears to be due to the gluten forming an indigestible tough inside both crop and gut (Bolton 1963).

3. *Barley.* This grain contains relatively high amount of N.F.E. (79.46 per cent) moderate content of crude protein (9.89 per cent), low content of crude fat (2.84 per cent) and the crude fibre content is 5.05 per cent.

The composition is similar to that obtained by the previous workers (Bolton 1963, Ghoneim 1958, MSz 6830T/65 I.v., Titus 1961).

The digestion coefficients of its nutrients (except the crude fibre) are of a high order, being 81.55 per cent, 79.06 per cent and 73.47 per cent for crude fat, N.F.E. and crude protein, respectively, similar to data obtained by Titus (1961) were 65.20 S. V. and 67.0 T.D.N.

The metabolizable energy of the grain is 1100 Kcal/lb (489 Kcal/kg) similar to that obtained by Bolton (1963).

The feeding value is lower than that found by other workers (Ghoneim 1958, MSz. 6830T/65 I.v.), when sheep was used to perform digestibility trials.

It contains 6.52 digestible protein.

The barley grain is excellent foodstuff for poultry and is used extensive than this. It is not so preferable as maize or wheat unless it is fed in an early age. Some poultrymen believe that better feather growth is obtained by feeding barley than by feeding maize and it is of some value to prevent cannibalism (TITUS 1961). When used in masses it should be finely and evenly ground.

4. Soya bean meal. The meal contains high amount of protein (43.07 per cent) being of leguminous origin. By solvent extraction its fat content is diminished to (1.88 per cent). It contains 6.63 per cent crude fibre and 42.46 per cent N.F.E. There is certain agreement between the composition of this sample and the data published by TITUS (1961), particularly when crude fibre, crude protein and crude fat are concerned, although the N.F.E. is lower.

During the experiment it was noticed that the daily faeces excreted by animals is too much: 322 g averagely, appr. 76.0 g dry matter (Ca. 67.0 g in dry matter basis).

The digestion coefficients of its nutritive components are not high, 72.66 per cent for crude protein, 49.26 per cent for the crude fat, and too low for the N.F.E. and crude fibre, 1604 and 10.69 per cent respectively. The feeding value is found to be 33.43 S. V. (36.74 T.D.N.).

It contains 28.1 digestible protein. The meal is considered to be rich in protein. It could be used to balance rations poor in digestible protein.

The feeding value obtained here is noticeably lower, than that recorded by other workers (BOLTON 1963, GHONIEM 1958, MSz 6830T/65, TITUS 1961).

5. Cotton seed meal. The sample of the cotton seed meal used here contained a low content of protein (19.88 per cent) crude fat (1.76 per cent) and crude fibre (10.68 per cent). It contained 61.0 per cent N.F.E., its composition differed from that indicated by TITUS (1961).

The digestion coefficients of its nutrients (except N.F.E.) are similar to that obtained by TITUS (1961), about 69 per cent, 83 per cent and 14 per cent for crude protein, crude fat and crude fibre, respectively. For N.F.E., it is too low (57 per cent).

The feeding value is 46 S. V. (49.42 T.D.N.). This value is in accord with that obtained by GHONEIM (1958) for the undecorticated cotton seed cake when used sheep to perform digestibility trial. On the other hand, this coefficient is low compared those obtained by other workers (BOLTON 1963, TITUS 1961).

It contains (12.60 per cent) digestible protein.

The fresh kernels contain two toxic principles, gossypol and gossypurpurin. These are destroyed by cooking the seeds in steam before the oil is removed. When unheated meal is fed in amounts too low to produce the toxic symptoms, yolk colour is adversely affected and for this reason it is not used much in feeding laying chickens.

6. Peanut meal. The analysis of this meal reveals high amounts of crude protein (44.62 per cent) and a moderate content of crude fibre (15.66 per cent). It contains low amounts of N.F.E. (30.35 per cent) and crude fat (2.79 per cent). This composition (except crude fat) is similar to that published by TITUS (1961).

The digestion coefficients of its nutrients are of high order for crude fat and crude protein (about 79 per cent and 74 per cent, respectively) and similar to those published by TITUS (1961) for the N.F.E. (it is about 60 per cent, but it is of low order for crude fibre about 18 per cent).

The feeding value is 47.50 S.V. (53.69 T.D.N.) .

Since the meal contains 30.09 per cent digestible protein it is a favourable source of protein and serves as good supplement to cereal grains. TITUS (1961) mentioned this meal as an excellent source of arginine but inferior to other plant protein supplements as source of methionine. BOLTON (1963) showed its choline content.

7. *Wheat bran.* The composition of the wheat bran is similar to that published previously (BOLTON 1963, GHONEIM 1958, MSz. 6830T/65, TITUS 1961). It contains relatively high amounts of crude protein (16.06 per cent) and N.F.E. (62.52 per cent), besides of 6.33 per cent crude fat and 8.80 per cent crude fibre.

The digestion coefficients of its nutrients are of high order for crude fat (86.46 per cent) and crude protein (80.69 per cent), but lower for N.F.E. and crude fibre (35.63 per cent and 20.13 per cent, respectively). The digestion coefficients obtained here (except N.F.E.) are higher than those obtained by TITUS (1961).

The feeding value is 36.76 S.V. (43.61 T.D.N.), similar to those calculated from the data of TITUS (34.36 S.V. and 41.0 T.D.N.). The value of the wheat bran obtained by BOLTON (1963) as metabolizable energy was 920 Kcal/lb (409 Kcal/kg), higher than resulted here (618 Kcal/lb 275 Kcal/kg).

Considering the feeding values obtained by GHONEIM (1958) using sheep in digestibility trials, his values were high, 72 S.V. for fine and 47 for coarse wheat bran.

It contains 11.46 per cent digestible protein.

This has rather low digestibility but is used to improve the appetite. It has a somewhat higher content of most of the required amino acides. TITUS (1961) mentions that wheat-bran had a rather high content of phosphorus, but most of it is not readily available. It is an excellent source of manganese.

8. *Lucerna dried meal.* This product is obtained from the grinding of the dry lucerne without any addition. Its value depends on its content of protein and fibre. The sample of this product used here contained moderate crude protein (17.15 per cent) and crude fibre (24.83 per cent), and low amounts of N.E.F. (48.31 per cent) and crude fat (2.35 per cent).

The digestion coefficients of its nutrients are high for crude fat (about 98 per cent) and crude protein (about 79 per cent), and low for N.F.E. (about 38 per cent) and crude fibre (29 per cent).

The feeding value is 25.70 S.V. as fed (40.20 T.D.N.) being similar to the values obtained by TITUS (1961) for the alfalfa meal, which were 24.79 and 36.0 for the S.V. and T.D.N.

Table 1

Analysis, digestion coefficients and feeding value of foodstuffs

Feedstuff	Dry matter as offered %	Composition of dry matter crude protein %	crude fat %	crude fibre %	N.F.E. %	Ash %
Maize	87.95	11.30	4.64	2.44	76.08	5.54
Wheat	86.25	11.73	2.41	1.95	81.18	2.73
Barley	89.37	9.89	2.84	5.05	79.46	2.76
Soya bean meal	89.80	43.07	1.88	6.63	42.46	5.96
Cotton seed meal	92.50	19.88	1.76	10.68	61.00	6.68
Peanut meal	91.06	44.62	2.79	15.66	30.35	6.58
Wheat bran	88.40	16.06	6.33	8.80	62.52	6.29
Lucerne dried	90.92	17.15	2.35	24.83	48.31	7.36

Digestion	Feedstuff		Coefficients		Feeding value	
	crude protein %	crude fat %	crude fibre %	N.F.E. %	starch value	T.D.N.
Maize	83.19	82.30	25.56	84.94	71.65	73.21
Wheat	84.64	69.56	18.59	84.89	70.38	71.51
Barley	73.47	81.55	13.18	79.06	65.84	67.81
Soyabean meal	72.66	49.26	10.69	16.04	33.43	36.74
Cottonsead meal	68.54	89.65	13.83	57.44	46.00	49.42
Peanut meal	74.05	79.12	18.13	59.69	47.50	53.69
Wheat bran	80.69	86.46	20.13	35.63	36.76	43.61
Lucerne dried	78.74	98.44	28.63	38.27	25.70	40.20

The metabolizable energy is 435 Kcal/lb (193 Kcal/kg), similar to that published by BOLTON (1963).

Comparing the value of the lucerne to the value of clover hay obtained by the author (DARWISH 1963) using sheep in digestibility trial, the starch value of the clover hay is slightly higher being 30.86 as fed (45.24 T.D.N.).

It contains 12.28 per cent digestible protein as fed. Although the lucerne dried supplies less total digestible nutrients than bran or any other dry foodstuff commonly used in feeding poultry, it is an extremely valuable ingredient of feeding formulae for chickens. Its value lies in its abundant vitamin content.

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DISTRIBUTION OF NON-SYMBIOTIC NITROGEN FIXING ORGANISMS IN SOILS OF LONG-TERM FERTILIZER TRIALS AND ROTATION EXPERIMENTS

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Non-symbiotic nitrogen fixing organisms namely *Azotobacter* and *Clostridia* have been counted in soil samples collected from the various plots of The Permanent Fertilization Experiment at Bahteem. *Azotobacter* has been found to be in higher densities than *Clostridia*. Organic manuring has significantly increased both *Azotobacter* and *Clostridial* populations, and capacity of nitrogen fixation much more than a complete mineral fertilizer. The rotation system seems to have no effect on such activities, however, the third year rotation plots showed somewhat higher activities.

Introduction

Non-symbiotic nitrogen fixing organisms are distributed all over the world (SUSHKINA 1949). Many investigators have, however, drawn the attention to the presence of high densities of such organisms especially *Azotobacter* in Egyptian soils, (ISHAC 1958), (MOBAREK 1960), (ABDEL-HAFEZ 1962), (ABD-EL-MALEK–ISHAC 1962), (VANCURA *et al.* 1962), (TAHA *et al.* 1965) and (MAHMOUD–TAHA-IBRAHIM 1968).

The present investigation entails the distribution of non-symbiotic nitrogen fixing organisms namely, *Azotobacter* and *Clostridia* in the different plots of the permanent fertilization experiment at Bahteem. This will show the effect of organic and inorganic fertilizers, in addition to the system of cultivation on non-symbiotic nitrogen fixation. This is well known to be of great benifit to soil fertility. Fixation of nitrogen as microbial protein is, however, the cheapest means of soil manuring.

Material and Method

Representative soil samples were collected from the different plots (0–20 cm) of the permanent fertilization experiment at Bahteem. This experiment has been established by the Egyptian Agricultural Society since 1912, to determine the effect of continuous application of fertilizers on the yields of crops. The experiment comprises the three rotations namely, the 1st year rotation (cotton), the 2nd year rotation (wheat–corn–cotton), and the 3rd year rotation (wheat–corn–cotton–clover). The fertilizer treatments were as follows:

1–0, control, received no fertilizers
2–N, received nitrate of soda.
3–NP, received nitrate of soda + superphosphate.
4–NPK, received nitrate of soda + superphosphate + sulphate of potash.
5–FYM, received farm-yard manure.

The percentages of organic matter, total nitrogen and pH values in the various plots are shown in Table 1.

Table 1

Some chemical properties of the permanent fertilization experiment at Bahteem

Treatments	1st year rotation			2nd year rotation			3rd year rotation		
	Org. matter %	Total N %	pH	Org. matter %	Total N %	pH	Org. matter %	Total N %	pH
O	1.15	0.074	8.2	1.08	0.070	8.2	1.06	0.070	8.1
N	1.22	0.076	8.4	1.16	0.075	8.5	1.10	0.078	8.3
NP	1.20	0.076	8.4	1.16	0.077	8.4	1.18	0.076	8.4
NPK	1.20	0.075	8.4	1.17	0.072	8.3	1.14	0.077	8.4
FYM	2.48	0.158	7.6	2.50	0.150	7.8	2.55	0.150	7.6

The soil samples were examined bacteriologically within 24 hours from sampling, after which they were air dried, ground and sieved through a 2 mm sieve and kept in glass bottles.

Azotobacter was counted on Base medium 77, while nitrogen fixing Clostridia was counted on modified Winogradsky's medium (ALLEN 1961). HOSKINS' tables (1934) were used for calculating the most probable numbers of these microorganisms, on oven dry basis.

To determine the capacity of soils for nitrogen fixation, 100 g of the air dried soil from each treatment was thoroughly mixed with 2 g glucose as a source of carbon and placed in petri dish in four replicates. The moisture content was continuously adjusted to 60 per cent of the water holding capacity. The plates were kept at 28° C for 1 month, after which carbon and nitrogen determinations were carried out.

Total nitrogen was determined by the modified Kjeldahl method, and organic matter was determined by the Walkely and Black wet digestion method (JACKSON 1958). pH values were determined in soil water suspension (1 : 5) with a Bekman glass electrode.

Results

Azotobacter and *Clostridia* counts are shown in Table 2. In general, *Azotobacter* was found to be present in high densities in all plots. This, however, induces us to stress their importance in nitrogen fixation in Egyptian soils.

Table 2

Counts of Azotobacter and Clostridia in Bahteem plots
Average numbers in millions/g dry wt. of soil

Treatments	1st year rotation		2nd year rotation		3rd year rotation	
	Azotobacter	Clostridia	Azotobacter	Clostridia	Azotobacter	Clostridia
O	5.3	2.4	7.0	2.8	8.4	1.9
N	3.9	4.0	5.0	3.2	9.9	4.2
NP	9.1	4.1	14.8	4.3	18.7	2.7
NPK	14.8	5.0	15.3	3.9	16.4	5.8
FYM	38.8	9.8	44.9	11.2	58.4	15.7

Organic manuring showed the highest count of *Azotobacter*, followed by treatments receiving NPK and NP, which showed little differences in counts denoting that potassium had little influence on such organisms. The figures obtained in NP and NPK treatments were, however, significantly higher than those obtained in N treatment. This shows the stimulating effect of phosphorus on the growth of *Azotobacter*.

Table 3

Nitrogen fixing capacities of Bahteem plots
Average g nitrogen fixed/100 g carbon oxidized

Treatments	1st year rotation	2nd year rotation	3rd year rotation
O	0.63	0.63	0.59
N	0.59	0.61	0.59
NP	0.60	0.66	0.97
NPK	0.76	0.96	1.10
FYM	1.66	1.66	2.35

The system of cultivation showed no significant effect on *Azotobacter* population as compared to the effect of fertilizers. However, the 3rd year rotation plots showed the highest count. This could be attributed to the rhizosphere effect of the different cultivated plants. Great stimulation of *Azotobacter* development in the rhizosphere of leguminous plants had already been noted by Löhnis (1928).

Counts of nitrogen fixing *Clostridia* showed the same trend of *Azotobacter*. No significant differences were recorded between the three rotations. The highest count of *Clostridia* was recorded in plots receiving organic manure. These were succeeded by those receiving inorganic fertilizers.

In the second part of this investigation, it has been found of interest to study the effect of the aforementioned treatments on the capacity of soils to fix the atmospheric nitrogen. Recently, soil biologists have drawn the attention to the microbial activities in the soil rather than the counts of microorganisms. Results are shown in Table 3.

The present data show clearly the significant effect of organic manure in promoting non-symbiotic nitrogen fixation. The relatively high amounts of fixed nitrogen recorded in plots receiving phosphorus and potassium besides nitrogen showed the importance of these elements in nitrogen fixation process. Phosphorus is well known to be essential element in the synthesis of protein and nucleoprotein.

The low figure recorded as a result of prolonged use of nitrogen could be explained by the fact that nitrogen fixers depended on soil nitrogen and became inactive in nitrogen fixation, since they showed higher counts in plots receiving nitrogen alone than the control plots. JENSEN (1940) stated that the amount of fixed nitrogen was proportional to the growth of *Azotobacter*. This, however, could be valid for the treatments receiving organic manure and to small extent for those receiving inorganic nitrogenous fertilizers. In the latter case, nitrogen fixers may seek the easiest way by obtaining nitrogen from the soil. Therefore, their counts could not be taken as an index of the assimilated nitrogen. Still, higher counts are significant since nitrogen will be bound as microbial protein rather than leaching in the soil. Such protein when mineralized will supply plants with available nitrogen (MAHMOUD—TAHA-IBRAHIM 1968).

Discussion

Egyptian soils are very interesting as regards the occurrence of non-symbiotic nitrogen fixing organisms. *Azotobacter chroococcum* occurs in the soils of Nile Valley in higher numbers than observed anywhere else (ABD-EL-MALEK—ISHAC 1962). They found more than one million per gram in 26 per cent of the soils tested and more than 10 million in 15 per cent of the soils. ALEXANDER (1961), however, stated that soils rarely contained more than 10 thousand organisms per gram.

Nitrogen fixing *Clostridia* was, however, found to be at low count as compared with *Azotobacter* in Egyptian soils (ABDEL-HAFEZ 1962, IBRAHIM 1964). On the other hand *Clostridia* was found to be at a higher density in soils of Europe (WAKSMAN—STARKEY 1949).

The process of nitrogen fixation was found by many investigators to be of great importance in Egyptian soils since they stated the presence of high densities of non-symbiotic nitrogen fixing organisms in such soils. Furthermore, VANCURA *et al.* (1965) added that the formation of important plant growth stimulators and vitamins by these organisms and their possible effect on soil fertility and plant growth is not to be overlooked.

Hence, it was found of interest in the present investigation to study the distribution of such organisms namely *Azotobacter* and *Clostridia* in the different plots of the Permanent Fertilization Experiment at Bahteem. Furthermore, this study will show the effect of continuous fertilization with certain organic (Farm-yard manure) and inorganic fertilizers (nitrate of soda, superphosphate and sulphate of potash) on the growth of such organisms and the capacity of soil to fix the atmospheric nitrogen. In addition, the effect of the system of rotation is also investigated.

The system of cultivation showed no significant effect as compared with fertilization on the growth and activities of non-symbiotic nitrogen fixing organisms. However, the third year rotation plots showed a somewhat higher activity regarding the counts of organisms and their activities over the 1st and 2nd year rotation plots. This could be attributed to the significant effect of the leguminous plant; to the clover usually cultivated in these plots.

The effect of fertilization was found to be more significant. Plots receiving organic manure showed the highest counts of microorganisms and the highest amounts of fixed nitrogen. This shows clearly the importance of organic matter from which Azotobacter could drive the necessary energy for growth and proliferation. JENSEN (1940) stated that the amount of fixed nitrogen was proportional to the growth of *Azotobacter*. This, however, could be valid in the present investigation for the soil receiving organic manure and to small extent for soil receiving inorganic nitrogenous fertilizer. In the latter case, *Azotobacter* may seek the easiest way by obtaining nitrogen from the soil. Therefore, their counts cannot be taken as an index of nitrogen fixation. However, higher counts are significant since nitrogen will be bound as microbial protein rather than leaching from the soil. Such protein when mineralized will supply plants with available nitrogen.

The high counts of *Clostridia* recorded in plots receiving organic manure could be deduced from several factors. The most important of them is the presence of high level of organic matter which can be fermented by *Clostridia*, as most of them are good fermentors of organic matter. Again, the presence of high counts of microflora (TAHA *et al.* 1965) is likely to cause association and symbiosis between the aerobic and anaerobic organisms. The former consumes oxygen furnishing suitable habitat for the latter to flourish.

Plots receiving phosphorus and potassium besides nitrogen showed a significant activity over those receiving nitrogen alone and the control. This could be deduced from the effect of these fertilizers in increasing crop yields (TAHA *et al.* (1965) and consequently crop residues. Such fertilizers also enhanced the decomposition of the unavailable organic matter. The application of potassium showed no significant effect in this respect. This could be explained by the fact that Egyptian soils are rich in this element (SERRY–MISHRIKI 1962).

WAKSMAN—STARKEY (1949), however, showed that the effect of inorganic fertilizers on soil microorganisms are exerted in a variety of ways: 1) Some constituents of the fertilizing materials may furnish a supply of elements which are deficient in the soil and may be consumed by the organisms. 2) The organic fertilizers may effect the physical properties of the soil, making it more or less favourable environment for the development of organisms. 3) They may affect the composition of soil solution by exerting solvent effects on the insoluble soil materials. 4) Salts may change the reaction of the soil solution. 5) Probably, the most pronounced effects upon microbes exerted by the addition of inorganic fertilizers are brought directly through higher plants, particularly where fertilizing practices have been in operation for extended periods of time; the response of plants to fertilizers is transferred to the soil organisms, as plant residues are incorporated in the soil.

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EFFECTS OF VARIOUS DOSES OF GAMMA IRRADIATION ON THE GROWTH OF PEA AND ON ITS PHOSPHATE AND SULPHUR METABOLISM

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In IP/1 pea plants irradiated with 0.5; 1.0; 5.0; 10.0 and 20.0 Kr doses the extent of inorganic 32-PO_4 uptake and incorporation in organic fractions increases in the third week following germination. Organic compounds of low molecular weight are exceptions when using irradiation with a dose of 5.0 Kr. Stimulation has a small-dose and a large-dose maximum. As regards incorporation of 35-SO_4 a similar tendency is shown with the difference that the large-dose maximum appears with 10.0 Kr applied, while with a dose of 20.0 Kr it decreases. On the other hand, irradiation (except for the 0.5 Kr treatment) has an inhibiting effect on the morphogenesis of plants; here too, a small-dose (1.0 Kr) and a large-dose (20.0 Kr) maximum of stem length can be observed.

Introduction

Plant species display different radiosensitivity as it is known from the respective literature (POLLHAMER 1965, BÁLINT—SUTKA 1965, SINGLETON 1958). Some authors attach importance to the ascorbic acid content of plants (VÁRTERÉSZ 1963), while others suggest that a quantitative determination of the nucleus may indicate by itself the radiosensitivity of the species examined (SPARROW *et al.* 1961, 1964, OSBORNE—LUNDEN 1964). Radiosensitivity can be modified by many other factors, too. So, when examining the sensitivity of a species it is just as important to take the developmental stage into consideration (BABAYAN *et al.* 1966), as to study the water content (KAPLAN 1939, 1949), the temperature and in connection with it the oxygen effect (NILAN—KONZAK 1961), the chemical factors (BÁLINT 1965) etc., as phenomena suggest that these factors induce changes by influencing the metabolic processes that determine the radiosensitivity.

The present paper tries to find an answer to the question: how changes the radiosensitivity of pea within a range of irradiation doses between 0.5 and 20.0 Kr, and what radiobiological changes are induced by the ionizing radiation taking into consideration the uptake and incorporation of $32-PO_4$ and $35-SO_4$.

Material and Method

The pea variety IP/1 was used in the experiments. Seeds with 54 per cent water content kept at room temperature were irradiated with a CO^{60} gamma radiation apparatus. The doses were: 0.5; 1.0; 5.0; 10.0; 20.0 Kr; in these treatments the margin of error was lower than ±10 per cent. Temperature during irradiation was 10° C; the duration of the treatment was three hours.

After the irradiation 100 seeds per treatment were germinated at room temperature in covered Petri-dishes out of which 10 per each were transplanted into pots after two days. The experiment was evaluated on processing of three-week-old pea plants, when, in addition to uptake and incorporation of 32-PO$_4$ and 35-SO$_4$, autoradiograms were made of leaves.

Uptake and incorporation of 32-PO$_4$ and 35-SO$_4$ were examined with Mothes' method (1954). Roots of plants removed from the pots were washed in tap water, then kept for 18 hours in a radioactive solution. Radioactivity of the solution was 0.5 mCi/100 ml for both isotopes. The two kinds of isotopes were contained in the same solution. Radiation of the 35-S was screened out by means of an absorbent layer of adequate thickness, and in the determination of the 32-P the effect of the absorbent was taken into consideration. When the time of incubation was up, the uppermost leaves were isolated, and the total uptake of radioactive 32-PO$_4$, the insoluble fraction as well, as the low molecular weight organic and inorganic phosphates soluble in 10 per cent tri-chloro-acetic acid were determined and expressed in Cpm. The same goes also to the sulphate content, except the soluble organic fraction of low molecular weight.

Results and Discussion

Uptake and incorporation of radioactive phosphates are presented in Table 1. Data shown in the tables are mean values of the per minute pulse numbers of the individual fractions and those of pulse numbers expressed in the percentage of the respective control. According to the data total anion uptake compared to the control increased with each radiation dose applied, but most intensively in case of the 0.5 Kr dose. Similar tendency was observed with the fractions, which are insoluble and soluble resp. in tri-chloro-acetic acid, and

Table 1

Uptake and incorporation of 32-PO$_4$ with various doses of irradiation applied

Treatment	Total		High molecular weight		Low molecular weight		Inorganic	
	Cpm	percentage of control	Cpm	percentage of control	Cpm	percentage of control	Cpm	percentage of control
Control	106.00	1.00	7.70	1.00	61.20	1.00	37.00	1.00
0.5 Kr	8506.00	80.30	1064.70	138.00	1710.00	27.90	5730.80	154.50
1.0 Kr	1064.10	10.50	91.40	11.80	561.30	9.16	411.50	11.08
5.0 Kr	546.50	5.15	55.80	7.25	51.70	0.84	439.00	11.80
10.0 Kr	760.60	7.22	76.50	9.94	616.40	10.05	67.80	1.83
20.0 Kr	1945.70	18.35	93.60	12.18	725.50	11.82	1779.60	48.00

Note: Results apply to fresh leaf weight, Cpm = imp/min/g × 10^2
deviation: ±10%

which contain inorganic phosphate (with the exception of the soluble organic fraction when applying 5.0 Kr); it was suggested that irradiation has a stimulative effect on the phosphate metabolism. There are, however, considerable differences in the extent of stimulation, since decrease observed with 5.0 and 10.0 Kr applied was followed repeatedly by an increased rate of phosphate uptake and incorporation in organic fractions when a dose of 20.0 Kr is applied. The critical minimum of the soluble organic fraction — including sugar phosphates, nucleotides, compounds with macroerg phosphoryl bonds — occurring in the 5.0 Kr treatment, indicates a lower than normal intensity of phosphorylation processes.

Concerning the uptake and incorporation of $35-SO_4$ presented in Table 2 (except the treatment with 20.0 Kr) the following can be established. Data on the incorporation of $35-SO_4$ should be considered to be of more universal validity than those on phosphate metabolism from the point of view of the intensity of protein synthesis. Uptake and incorporation of $35-SO_4$ increased significantly when applying 0.5; 1.0; 5.0 and 10.0 Kr doses respectively, but remained below the control in the case of 20.0 Kr. It must be mentioned that anion uptake expressed in the percentage of control was similar in the case of $32-PO_4$ and $35-SO_4$ as well.

Figures 1, 2 and 3 clearly show that at this developmental stage the irradiation — with the exception of doses of 20.0 and 5.0 Kr — has a stimulative effect on the metabolic part processes examined. This does not mean the similar development of the morphogenesis, on the contrary, latter shows in many cases an inverse tendency. It can be seen that the increased ion uptake induced by a radiation dose of 0.5 Kr results in plants larger than the control.

Table 2

Effect of ionizing radioactive irradiation on $35\text{-}SO_4$ uptake and incorporation in the protein fraction of pea

Treatment	Total		High molecular weight		Inorganic	
	Cpm	percentage of control	Cpm	percentage of control	Cpm	percentage of control
Control	262.5	1.00	9.8	1.00	211.1	1.00
0.5 Kr	15 291.2	58.20	68.9	7.04	15 209.8	71.85
1.0 Kr	2 492.0	9.47	77.9	7.85	1 052.0	4.97
5.0 Kr	1 171.4	4.46	30.8	3.14	1 115.5	5.27
10.0 Kr	1 619.5	6.16	36.8	3.76	1 576.4	7.45
20.0 Kr	209.0	0.79	9.0	0.93	166.6	0.78

Note: Results apply to fresh leaf weight, $\text{Cpm} = \text{imp/min/g} \times 10^2$
deviation: ±10%

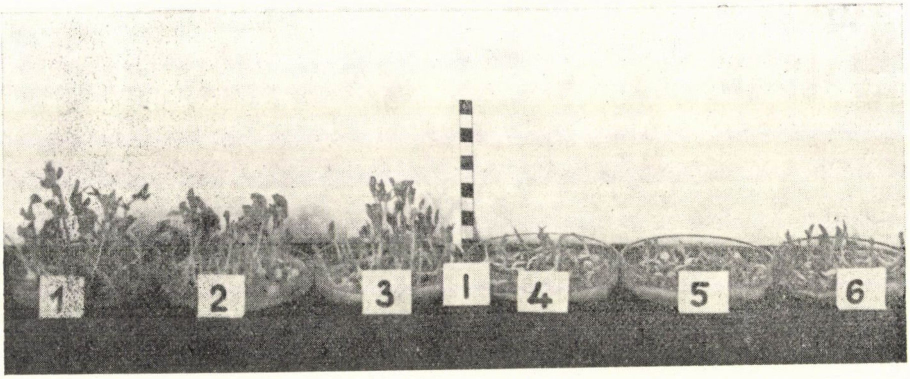

Fig. 1. Investigation of the effect of cobalt gamma irradiation on the fodder-peas on the 21st day after irradiation. 1 = control; 2 = 0.5 Kr; 3 = 1.0 Kr; 4 = 5.0 Kr; 5 = 10.0 Kr; 6 = 20.0 Kr

Fig. 2. Comparison of root system of fodder-peas on the 21st day after irradiation. 1 = control; 2 = 0.5 Kr; 3 = 1.0 Kr; 4 = 5.0 Kr; 5 = 10.0 Kr; 6 = 20.0 Kr

Thus the stimulative dose seems to be around 0.5 Kr, doses higher than that result in considerable morphological changes: indented, wrinkled leaves, reduced leaf surface area — that is, in a depressed development and growth of the whole plant. The highest dose applied (20.0 Kr) caused — in addition to a slightly reduced total stem length — indented, wrinkled, mottled leaves.

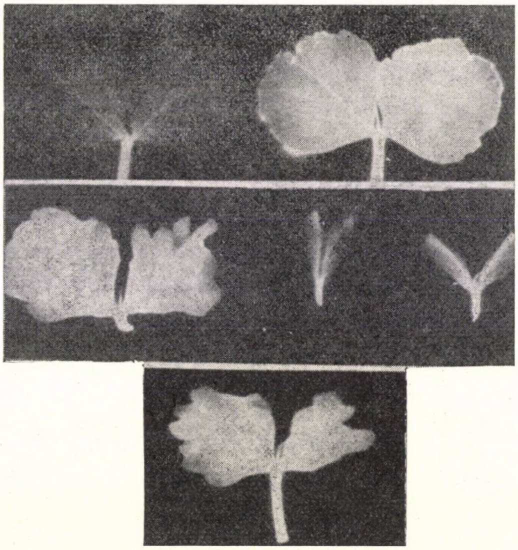

Fig. 3. Autoradiographic of pea-leaves on the 21st day after irradiation. 1 = control; 2 = = 0.5 Kr; 3 = 1.0 Kr; 4 = 5.0 Kr; 5 = 10.0 Kr; 6 = 20.0 Kr

Conclusions

When applying large doses of irradiation to peas Avakyan *et al.* (1967) observed a similar phenomenon as regards chromosome aberrations, respiration intensity and germination.

According to the data of experiments ionizing irradiation has an almost identical effect on the uptake and incorporation of $32-PO_4$ and $35-SO_4$, with the exception of the critical dose of 20.0 Kr, which decreases the uptake and incorporation in proteins of the $35-SO_4$ isotope to a great extent. The intensive stimulation observed with the dose of 5.0 Kr applied can be possibly explained by the reduced permeability of cell-walls, as this treatment enhanced the ion uptake of cells which, in turn, resulted in an increased metabolic activity. On the other hand, with higher — 5.0 and 10.0 Kr — doses applied, metabolic processes, which are less sensitive to irradiation, can be supposed to suffer. E.g. it is possible that the biosynthesis of a less radiosensitive metabolate has still a somewhat positive, stimulative effect as compared to the control, due to an increased ion uptake and supposedly increased cell respiration, while the synthesis of enzymes required for growth suffers considerably from irradiation.

It is a very interesting observation that an irradiation dose just below the lethal dose (20.0 Kr) has a lower growth inhibiting effect than doses of 5.0 and 10.0 Kr. The second maximum can be characterized by the increased intensity of phosphate metabolism as well. Action mechanism of ionizing irradiation similar to that of the antimetabolites has not, however, been cleared so far. It can be supposed, on the other hand, that an enzyme synthesis displaying earlier a relative minimum, later increases again, as all metabolic processes decrease in intensity. The lower maximum observed before the lethal dosis applied can be possibly explained by this.

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GROWTH PROMOTION SPECIFICITY EXHIBITED BY AQUEOUS EXTRACT OF GALLS IN SALVADORA PERSICA L.

By

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Salvadora persica is a desert plant which forms galls in almost any part. These galls have been reported here to possess not only growth-inhibiting but also prominant growth-promoting substances. The influence of the promoting action of the aqueous extract from the galls has been specifically different on the growth of the two species *(Pennisetum typhoideum* and *Asteracantha longifolia)* selected for the present study. There appeared to be more than one growth-promoting substances as indicated by their different Rf values, but only a few were noteworthy. These growth-promoting substances were thermostable as the autoclaving of the extract did not affect their quality, whereas the growth-inhibiting substances appeared to be thermolabile.

Introduction

Salvadora persica is a common plant in the Indian desert. Galls are commonly formed on any part of this plant. In fact, it is due to the presence of some unidentified insects inside the galls which results in the formation of this abnormal cancerous growth. The galls do not possess any particular shape (Fig. 1).

There are a number of reports about the occurrence of inhibitors in fruits and seeds (EVENARI 1949). Hardly any report regarding growth-promoting substances exists in the literature. SIRCAR *et al.* (1959, 1960) have reported on growth-regulating properties in the root extract of water hyacinth. KATHJU *et al.* (1969) reported on the presence of growth-inhibiting substances from the different parts of *S. persica* like leaf, stem and bark extracts, except for the aqueous extract of dry stem which promoted growth of *Cyamopsis tetragonoloba.* SEN *et al.* (1969) have reported not only on growth-inhibiting but also on growth promoting principles in the stem of *Prosopis juliflora* for the germination and growth of *Euphorbia caducifolia* seeds.

The present work has been undertaken to find the reason of this tumorous growth by both cell enlargement and cell division in the galls. It was presumed that some substances causing this growth may be present there, the effect of which could be analysed. The present study reports on the detection of a few growth promoters in the galls.

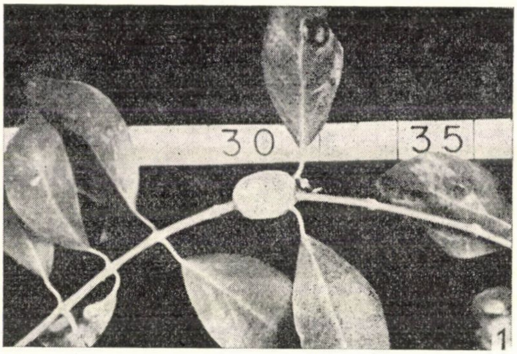

Fig. 1. Salvadora persica branch showing the nature of galls

Material and Method

Galls were collected from different parts mainly from young branches. Weighed material was crushed and extracted in a known quantity of distilled water (25 gm material in 100 ml of distilled water). Before filtering, the crushed material was allowed to stand as such overnight, so that all possible substances present in the galls may diffuse into the water. The clear solution after filtering was diluted to different concentrations (1 per cent; 2.5 per cent; 5 per cent and 10 per cent). A part of all the dilutions used was autoclaved to see the stability of the substances against heat.

Effect of the extract was observed on the germination of seeds and seedling growth of *Asteracantha longifolia*, a weed and *Pennisetum typhoideum*, a cultivated crop plant. Extract from the galls was also prepared by direct centrifugation of the chopped galls without crushing and also not allowing the extract to stand overnight.

Chromatograms were also run using acetic acid and water as solvent to find the Rf values of the substances affecting growth in both unautoclaved and autoclaved extracts.

Results

Effect of aqueous extract obtained by crushing. The seeds of *A. longifolia* and *P. typhoideum* were germinated in the various dilutions of the extracts as mentioned above and also in distilled water for control. There has been almost no effect on germination of seeds of either of the two above-mentioned species. But the influence on growth of seedlings has been remarkable. The results obtained have been incorporated in Tables 1 and 2.

Pennisetum typhoideum. From Table 1 it would become evident that autoclaving of the extracts somehow enhanced the promoting effect on growth of seedlings. It might be presumed that the reported growth —inhibiting principles in *S. perdica* were thermolabile and the effect of the latter got prominance after autoclaving of the extracts. In case of *P. typhoideum* root growth was inhibited in unautoclaved extracts, whereas in autoclaved one its growth was almost twice as much as that of the control in 1 per cent to 5 per cent concentrations, so much that even in 10 per cent extract the growth was much more

Table 1

Effect of different concentrations of unautoclaved and autoclaved extracts from the galls of Salvadora persica on the seedling growth of Pennisetum typhoideum
(Three-day-old seedlings)

Concentration of extracts	Unautoclaved extract		Autoclaved extract	
	Radicle in mm	Shoot in mm	Radicle in mm	Shoot in mm
Control	47.3 ± 18.8	32.0 ± 7.4	22.2 ± 8.2	23.6 ± 3.8
1 per cent	36.4 ± 14.4	32.7 ± 6.7	40.6 ± 14.9	38.3 ± 5.8
2.5 per cent	30.7 ± 2.2	39.7 ± 5.1	40.5 ± 8.5	40.1 ± 6.4
5 per cent	30.3 ± 9.0	36.2 ± 6.3	39.2 ± 8.7	42.6 ± 5.4
10 per cent	6.6 ± 2.1	22.0 ± 3.0	32.5 ± 11.6	38.5 ± 8.6

Table 2

Effect of different concentrations of unautoclaved and autoclaved extracts from the galls of Salvadora persica on the seedling growth of Asteracantha longifolia
(Five-day-old seedlings)

Concentration of extracts	Unautoclaved extract		Autoclaved extract	
	Radicle in mm	Hypocotyl in mm	Radicle in mm	Hypocotyl in mm
Control	19.3 ± 5.4	13.3 ± 1.1	25.4 ± 5.0	14.2 ± 1.9
1 per cent	28.7 ± 4.8	18.0 ± 1.9	35.3 ± 11.6	20.2 ± 4.7
2.5 per cent	33.2 ± 7.0	22.6 ± 5.0	28.6 ± 5.0	21.4 ± 4.1
5 per cent	24.6 ± 5.6	18.8 ± 3.5	26.1 ± 4.2	23.0 ± 3.3
10 per cent	12.0 ± 5.0	11.2 ± 3.6	18.0 ± 7.1	16.0 ± 3.6

than the control. Shoot growth also exhibited a little inhibiting effect in unautoclaved extracts but in high concentration of 10 per cent, although the promoting effect in autoclaved extracts has been tremendous. In both unautoclaved and autoclaved material samples of 5 per cent concentration exhibited the best growth of shoot and at the same time did not differ very much from each other. It is interesting to note that the shoot remained unaffected by the inhibitory effect of the extracts. The effect on root growth may also be toxic as it remained in contact with the extract whereas the shoot did not. Ten per cent extract has inhibited growth of shoot in unautoclaved extract, but it is promoting in autoclaved ones. It might be concluded that in unautoclaved extracts promoting principles appear on dilution when the inhibitory ones remain ineffective. This may be compared with many growth-promoting substances which remain so when applied in low concentrations, but their effect becomes inhibitory in high concentrations (Fig. 2).

Asteracantha longifolia. It would be evident from Table 2 that radicle as well as hypocotyl are affected beneficially by autoclaved extracts of *S. persica* galls in case of *A. longifolia*. The unautoclaved extract exhibited promotion of growth in radicle and hypocotyl till 5 per cent concentration. In unautoclaved

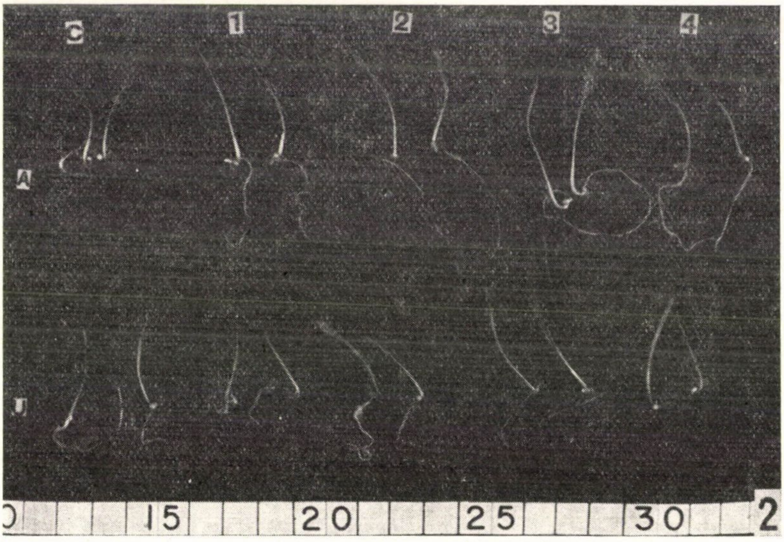

Fig. 2. Effect of aqueous extract both autoclaved (A) and unautoclaved (U) from the galls of *S. persica* on the growth of *P. typhoideum* seedlings (three days old) in different concentrations. C = Control; 1 = 1 per cent; 2 = 2.5 per cent; 3 = 5 per cent and 4 = 10 per cent

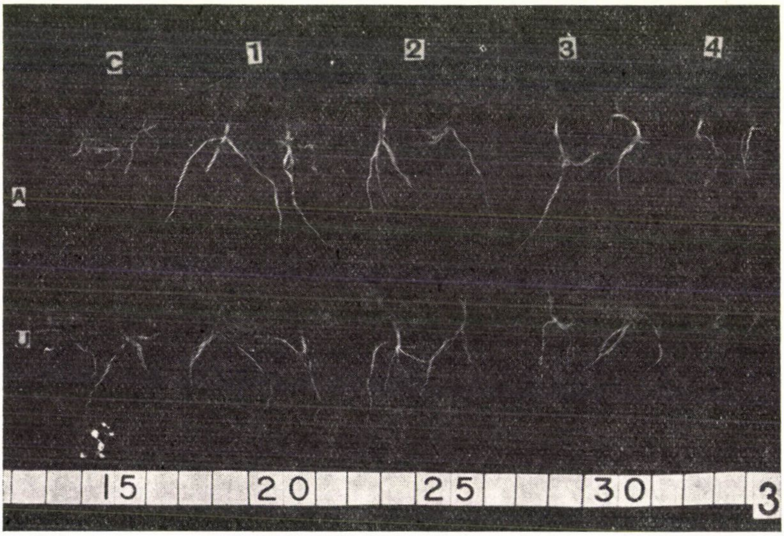

Fig. 3. Effect of aqueous extract both autoclaved (A) and unautoclaved (U) from the galls of *S. persica* on the growth of *A. longifolia* seedlings (five days old) in different concentrations. C = Control; 1 = 1 per cent; 2 = 2.5 per cent; 3 = 5 per cent and 4 = 10 per cent

extract the best root growth took place in 2.5 per cent extract whereas in autoclaved one it was in 1 per cent extract, thus again proving that promoting principles were more effective in dilute solutions. Unautoclaved extract of 2.5 per cent and the autoclaved one of 5 per cent concentration exhibited the best promoting effect on hypocotyl of *A. longifolia* seedlings. In autoclaved extract the promotion of hypocotyl was visible even in 10 per cent concentration (Fig. 3).

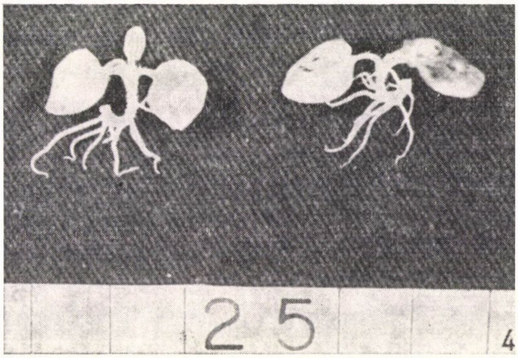

Fig. 4. Root initiation on hypocotyl as an effect of *S. persica* extract after decapitating the hypocotyl near the hypocotyl/radicle transition region in *A. longifolia* seedlings (after seven days)

Since there has been promoting effect on root growth, it was thought advisable to see the effect of *S. persica* gall extract on root initiation and their growth after decapitating the hypocotyl near the hypocotyl/radicle transition region. The rootless shoot was cultured in unautoclaved aqueous extracts of the above-mentioned four concentrations. It was seen that enormous number of adventitious roots emerged from all around the hypocotyl, especially from the portion which was in contact with the extract but not from the cut end (Fig. 4). The best concentrations of the extracts for root initiation and growth have been 1 per cent and 2.5 per cent as compared to the control. There have been extremely poor root formations in control and 10 per cent extract of the galls.

Effect of the extract obtained by centrifugal force. Crushing the galls and keeping them in water for a long period might tend to increase the unwanted substances in the extract. So infiltration of growth-promoting (inhibiting as well) principles were obtained by centrifugation for some time. The extract so obtained was diluted accordingly and the effect of growth observed. Root hairs were abundantly formed in roots not contaminated by extract as compared to the one dipped in the extract. The effect of the extract obtained by centrifugation is tabulated in Table 3.

Table 3 again indicates that the radicle in *P. typhoideum* was inhibited, but at the same time it was extremely clear that promoting principles present were far more even in high concentration than that of 10 per cent, the same was true for shoot as well, although the latter was promoted at 1 per cent concen-

Table 3

Effect of different concentrations of unautoclaved extract obtained by centrifugation from the chopped galls of Salvadora persica on the seedling growth of Pennisetum typhoideum (three days old) and Asteracantha longifolia (five days old)

Concentration of extracts	*Pennisetum typhoideum*		*Asteracantha longifolia*	
	Radicle in mm	Shoot in mm	Radicle in mm	Hypocoytl in mm
Control	47.3 ± 18.8	32.0 ± 7.4	19.3 ± 5.4	13.3 ± 1.1
1 per cent	23.4 ± 5.2	36.4 ± 5.9	21.7 ± 6.0	13.6 ± 3.5
2.5 per cent	37.6 ± 3.7	21.8 ± 11.1	25.5 ± 6.7	16.5 ± 3.3
5 per cent	37.0 ± 7.9	31.4 ± 8.4	20.6 ± 3.3	14.9 ± 4.4
10 per cent	43.2 ± 14.8	28.0 ± 7.4	11.1 ± 6.4	15.8 ± 3.3

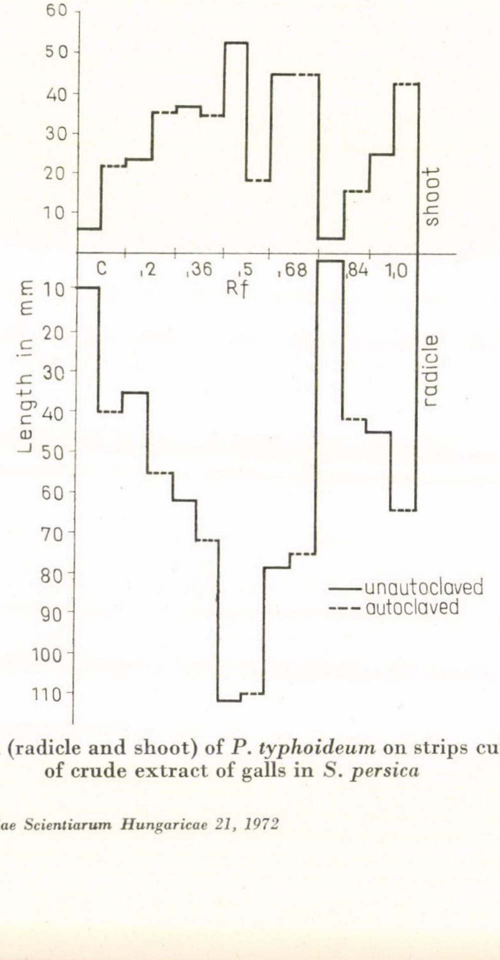

Fig. 5. Seedling growth (radicle and shoot) of *P. typhoideum* on strips cut from chromatograms of crude extract of galls in *S. persica*

tration. In case of *A. longifolia*, the radicle and hypocotyl promotion was prominent, although it was evident in hypocotyl even in higher concentrations as compared to radicle.

From this experiment it might be concluded that the infiltration of substances promoting growth were comparatively more but the dominance of inhibitory substances do not let their effect become prominent. It was probable that centrifugation for a longer time and with higher gravitational force, the result may be different.

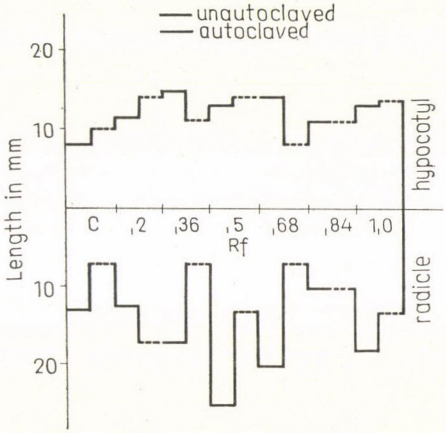

Fig. 6. Seedling growth (radicle and hypocotyl) of *A. longifolia* on strips cut from chromatograms of crude extract of galls in *S. persica*

Separation of the growth regulators. Both unautoclaved and autoclaved extracts of the galls were spotted on Whatman No. 1 filter paper for chromatography in 6 per cent acetic acid solvent system. Chromatograms were dried in air and each was cut into equal segments. Segments were separately eluted in equal volume of distilled water by keeping each in a petri dish and the seeds were germinated for observing the effect on growth.

On chromatograms of crude unautoclaved extract an inhibitor was found at Rf ·84 affecting both radicle and shoot growth of *P. typhoideum*, whereas strong promoter was evident in both unautoclaved and autoclaved ones at Rf ·5 except for affect on shoot in the latter extract, which showed promoting effect at Rf ·68 (Fig. 5).

In case of *A. longifolia*, the chromatograms of unautoclaved crude extract indicated a promoter affecting the radicle and shoot growth at Rf ·36 and ·5 respectively, whereas in autoclaved one the promoter was evident both for radicle and shoot at Rf ·2 and also at ·5 for only shoot growth (Fig. 6).

Discussion

The presence of growth inhibitors has been reported in desert plants. Parts of *S. persica*, on the galls of which the present study is based, being said to have growth-regulators are reported in different parts (dry leaves, dry twigs, fresh and dry inner cortex) of this species as tested against germination and growth of *Cyamopsis tetragonoloba* (KATHJU *et al.* 1969).

As compared to the inhibitors, the knowledge of promoters would be more beneficial for the afforestated purposes and improvement of desert. SEN *et al.* (1969) have reported on the presence of growth-promoting principle(s) in the stem of *Prosopis juliflora* which is also a desert plant. Similarly KATHJU *et al.* (1969) have found that the aqueous extract of dry stem of *S. persica* promoted the root growth of *C. tetragonoloba*. The dominance of some growth promoting substance in the galls of *S. persica* is very much noteworthy.

The aqueous extract from the galls of *S. persica* showed a differential specificity when tested against the growth of *P. typhoideum* and *A. longifolia* seedlings as regards its promoting effect. There was absolutely no effect on germination but the effect on growth was prevalent. The growth-promoting substances appeared to be thermostable as its effect became more prominant in the autoclaved extracts, apparently because the growth-inhibiting substances turned out to be thermolabile. The thermolabile nature of the growth-inhibiting substance has also been reported by KATHJU *et al.* (1969). The growth-promoting specificity exhibited by the gall extract was different for the two plant species tested. The radicle was inhibited in *P. typhoideum* but was promoted in *A. longifolia* till 5 per cent concentration, although the effect on shoot of both the species was promoting to a dilutation of 5 per cent in unautoclaved extracts. The autoclaved extracts promoted the root and shoot growth in both the species but for the former in *A. longifolia* to a certain extent. The promotion of root growth in 1 per cent and that of shoot in 5 per cent concentration took place. On *A. longifolia* seedlings the effect of unautoclaved and autoclaved extracts was beneficial, although the magnitude of growth differed in different concetrations. The presence of root-promoting substance was further confirmed by culturing the decapitated plant from the radicle/hypocotyl transition region, where root formation took place abundantly in *A. longifolia*.

Extraction of growth-regulating substance by centrifugation in water at high gravitational force and for a considerably long time would let more promoting substances to infiltrate in water as compared to crushing when all cells are injured and produce more toxic effect on growth.

The growth-promoting substances in unautoclaved extracts have been effective in low concentrations whereas they were so in higher ones as well in autoclaved extracts. It was observed that Rf values of the promoting substances varied for the two species as also for radicle and shoot indicating that there may be several growth-promoting substances. The chemical properties and other physiological effects of the growth regulators in *S. persica* galls as compared to known growth regulators are yet to be analyzed.

Acknowledgements

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STUDY ON THE RELATION OF GERMINATION INABILITY AND DEHYDROGENASE ENZYME ACTIVITIES IN PEA SEEDS

By

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Studying the cause of germination inability from the aspect of sufficient or insufficient activities of enzymes catalizing the germination physiological processes, the paper finds a correlation between extractable, extractable + bound and topochemically determined dehydrogenase activities of the seed on one hand, and percentage germinating capacity expressing the ratio of germinating and non-germinating seeds, on the other.

Introduction

Several authors find relations between germinating capacity and its demonstration, and activities of dehydrogenase enzymes contained in the seed (LAKON 1939, 1942, 1949, 1950, 1953, MORALES 1963, PLAUT–HALFON 1954).

The topographical determination of germinating capacity (viability) introduced by Lakon consists of precipitating water insoluble, red formazane on seed tissues placed in diluted 2,3,5-triphenyl-tetrazolium-chloride and displaying dehydrogenase activity while the inactive places remain white. On the basis of the colouring the germinating capacity can be established according to Fig. 1. In this context LINDNER (1955) points out that topographical germinating capacity determination recommended by Lakon is based on the supposition that the dehydrogenase activity of germ-cells corresponds to the germinating capacity. According to his investigations, under special influence (drying, seed dressing, hot water treatment, etc.) germinating capacity of seeds decreased much more rapidly than the dehydrogenase activity. From this observation Lindner draw the conclusion that in these cases cell activities were so much disturbed that the TTC (2,3,5-triphenyl-tetrazolium-chloride) test could not be used for the evaluation of germinating capacity.

MACLEOD (1952) examined the germinating capacity and enzyme activities of barley as a function of age. With advanced years germinating capacity decreased the most rapidly while dehydrogenase activity could be demonstrated even when germinating capacity fell to zero.

The question arises how close is the relation between germinating capacity and dehydrogenase activity? Is there any difference according to whether dehydrogenases are determined topochemically or studied independent of their place of action?

Can a closer correlation be disclosed with the germinating capacity according to whether it is dehydrogenase activity in general, or as related to substrates contained by the seed, or else the activity of a special enzyme that is studied?

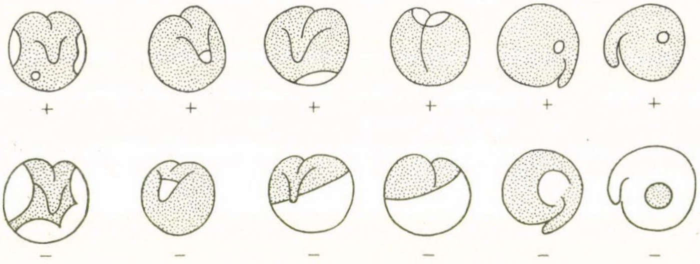

Fig. 1. Colouring of seeds able and unable to germinate respectively, in the course of TTC treatment (Lakon 1953) (+able to germinate, — unable to germinate)

It is important to answer these questions because a close correlation — if found — between dehydrogenase activity of seeds and germinating capacity would provide a possibility to approach the causes of enzyme induced germination inability, as percentage germinating capacity expresses the ratio of germinating and non-germinating seeds. In case of a close correlation any change in this ratio should be reflected by a changed dehydrogenase activity. Thus, from the extent of dehydrogenase activity displayed by a sample of low percentage germinating capacity (i.e. where non-germinating seeds prevail) and the extent of dehydrogenase activities found in other samples of higher percentage germinating capacity conclusions can be drawn — in case of a close correlation — on germination inability. No direct method of determining germination inability is available, since it is only after having been germinated that seeds turn out to be either able or unable to germinate, and, when dry — i.e. in advance — cannot be separated into germinating and non-germinating seeds.

Peas are sometimes harvested with a very low percentage germinating capacity; its unfavourable economic effects make it necessary to solve the problem by finding the causes of germination inability as soon as possible.

Material and Method

In one-year old pea samples of different variety and germinating capacity extractable as well as extractable + bound dehydrogenases of dry, ground seed samples on one hand, and places of seed surface showing dehydrogenase activity, on the other, were examined as functions of percentage germinating capacity.

Extractable degydrogenases were examined with Thunberg's methylene blue technique. On one hand dehydrogenase activity of the seed extract with its own substrates, on the other succinic acid-, isocitric acid-, and malic acid-dehydrogenase activities were determined, where substrates (succinic acid, citric acid, malic acid and their potassic salts respectively) added to the enzyme extract represented the hydrogen donor, and methylene blue the hydrogen acceptor which — with the intervention of dehydrogenases was reduced to leucomethylene blue. The speed at and degree to which the blue colour disappears indicate the extent of the dehydrogenase activity (BELOSERSKI—PROSKURYAKOV 1956). The rapidly reduced colour is easily reoxidized by the oxygen of the air, therefore these reactions can only be studied in specially made, evacuable "Thunberg recipients".

Before the examinations the seed-coat should be removed as, on one hand, it does not contain dehydrogenases, on the other, due to its exchange matters inactivates a great proportion of dehydrogenases. With 0.8 percent K_2HPO_4 dilution a 20 percent suspension was made of ground, coat-less seeds, then — after shaken for ten minutes — filtrated. The mixture obtained by 1 ml 0.1 mol substrate solution, 2 ml Sörensen buffer of pH 6 and 0.5 ml $2.67 \cdot 10^{-4}$ mol methylene blue solution added to 5 ml filtrate was evacuated in Thunberg's tube for 3 minutes, then placed in a water-bath of 37° C, and the time required for the disappearance of the blue colour measured. The sample inactivated by boiling keeps its blue colour. 1 ml 10^{-3} mol methylene blue absorbs 22.41 μl hydrogen. Dehydrogenase activity figure was calculated on this basis; it indicates the μl amount of hydrogen 1 g seed-coat free, air-dry ground pea seed transfers in an hour to the methylene blue through the activity of its dehydrogenases.

The relative error of measuring is: $m\% = \pm 3.150$.

The dehydrogenase activity with the own substrate of the seed was also measured, without enzyme extract being made. In this way extractable + bound dehydrogenase activities were measured together; their extent was indicated by the amount of the red formazane obtained with the use of TTC.

Coat-free ground pea seed was also used to determine the formazane production of pea samples of different germinating capacity 7 ml 1 per cent TTC solution was poured onto a 0.50 g amount of ground seed, and the suspension thus obtained shaken for 20 minutes at room temperature. The 1 per cent TTC solution was prepared with Sörensen buffer of 6.6 pH-value added. The surfaces of the samples were covered with a toluol layer to keep off the air, then the samples were incubated for 15 hours at 30° C. Seed flour was subsequently separated from the solution by filtration, and the red formazane produced extracted with acetone. Boiling accelerates the dissolution of formazane. The extract was diluted to a final volume of 100 ml, and its extinction measured in Pulfrich's photometer with S50 colour filter used. The amount of formazane corresponding to extinction values read was determined by means of a calibration curve. The calibration curve was drawn in the following way: different amounts of TTC were reduced (in N NaOH) with ascorbic acid, formazane precipitated dissolved with acetone, and extinction values of extracts diluted to the same final volume plotted against formazane amounts corresponding to the known amounts of TTC.

The activity figure is: mg formazane/1 g ground seed, that is, the mg amount of formazane produced by 1 g seed.

The relative error of measuring is: $m\% = \pm 0.914$.

Percentage germinating capacity was given by the number of seeds germinating in eight days at room temperature out of 100 pea seeds placed between wet layers of filter paper. Percentage germinating capacity was determined in each sample from the average of four replications.

Percentage germinating capacity was also determined topographically. For this purpose 2×100 pea seeds were placed in a 1 per cent solution of TTC solved in Sörensen fubber of pH 6.6 and incubated for 15 hours at a temperature of 30° C. Due to the dehydrogenase activity at places of active cells red formazane spots appeared, which were evaluated according to Table 1.

Results

Studying the causes of enzyme induced germination inability the relation between percentage germinating capacity and activities of dehydrogenases extracted (extractable) and not extracted (extractable+bound) from the seed,

Table 1

Malic acid, succinic acid and isocitric acid dehydrogenase activities of seed extracts, as functions of percentage germinating capacity, in three pea varieties
(r = correlation coefficient)

Variety	Germinating capacity (% of germinated seeds)	Succinic acid dehydrogenase activity	Isocitric acid dehydrogenase activity	Malic acid dehydrogenase activity
Victory Freezer	54	4.7	7.7	13.5
(Pisum sativum quadratum)	61	5.9	6.0	15.4
	65	2.7	3.6	12.0
	71	3.6	5.6	5.3
	74	3.1	5.4	15.4
		$r = -0.629$	$r = -0.565$	$r = -0.298$
Petit Provençal	47	0.5	2.2	3.8
(Pisum sativum pachylobum)	80	2.8	5.6	17.9
	90	2.7	3.6	4.7
		$r = +0.990$	$r = +0.657$	$r = +0.382$
Újmajori korai Viktória	56		3.1	6.7
(Pisum sativum pachylobum)	61	activity	2.9	5.7
	69	hardly	2.0	2.9
	75	measurable	2.6	3.6
	92		3.5	6.0
		$r = 0$	$r = +0.270$	$r = -0.148$

on the one hand, and places of seed surface showing topochemically demonstrable dehydrogenase activity on the other, were also examined.

Table 1 shows the relation of malic, succinic and isocitric acid-dehydrogenases extracted from the seed — i.e. extractable dehydrogenases — to the percentage germinating capacity.

The absolute values of correlation coefficients included in the table indicate the intensity of correlation while their signs the direct or inverse character of correlation.

The r-value reveals an equally loose relation between percentage germinating capacity and malic acid-dehydrogenase activity in all three varieties. There is a loose connection — now direct, now inverse according to the variety — between isocitric acid-dehydrogenase activity and germinating ability, except for the variety "Újmajori korai Viktória", which shows the least relation

Table 2

Extractable and extractable + bound dehydrogenase activity of pea samples of different germinating capacity as related to substrates contained in the seed (r = correlation coefficient)

Variety	Germinating capacity (% of seeds germinated)	Extractable dehydrogenase activity	Extractable + bound dehydrogenase activity
Victory Freezer	54	21.5	10.8
(Pisum sativum quadratum)	61	12.0	11.3
	65	17.9	10.4
	71	18.9	9.7
	74	21.5	6.6
		$r = -0.204$	$r = -0.774$
Újmajori korai Viktória	56	6.7	4.0
(Pisum sativum pachylobum)	61	8.9	4.6
	69	3.1	3.5
	75	5.4	4.1
	92	10.7	6.7
		$r = +0.399$	$r = +0.727$
Petit Provençal	47	3.6	2.7
(Pisum sativum pachylobum)	80	16.5	5.1
	90	6.7	4.0
		$r = +0.494$	$r = +0.764$

concerning all three enzymes examined in any case. And as for the succinic acid-dehydrogenase enzyme, in the variety Petit provençal, activity increases parallel with the percentage germinating capacity; according to the r-value there is a close direct relation between the two factors, in contrast with the variety Victory Freezer where this relation is inverse.

Table 2 shows the relation of extractable and extractable + bound dehydrogenase activities with the substrates of the seed to percentage germinating capacity. The table reveals that a closer relation can be found between germinating capacity and enzymes activities when dehydrogenases are not extracted from the seed before activity is measured — that is, when dehydrogenase (extractable + bound dehydrogenase) activities in the seed grists are measured — than when activity of the enzyme extract is determined.

When comparing the signs of r-values in Tables 1 and 2 from the point of view of varieties examined, we find a decreasing activity with increasing percentage germinating capacity in the variety Victory Freezer, independent of the character of dehydrogenases studied — if examined between sufficiently wide limits; i.e. there is an inverse, though not equally intensive relation between the two factors. When examining the variety Petit provençal in the same way, we find a direct, more or less intensive relation between the two factors, in other words an increasing percentage of germinating capacity means an increased dehydrogenase activity. The variety Újmajori also displays a direct relation, but here intensive relation can only be found in the case of extractable + bound dehydrogenase activity. On the bases of all this it can be established that the varieties examined differ in the character of relation between percentage germinating capacity and dehydrogenase activities; the marrowfat pea variety showed trends contrary to those displayed by the round seeded peas.

Table 3 shows a close correlation between germinating capacity determined by germination and that determined topographically. A comparison of the experimental results suggests that viability can be more correctly determined according to the number, size and location of dehydrogenase activity sites on the seed surface than by the extent of activity shown by the whole seed.

Table 3

Germinating capacity determined by germination and topographically
(r = correlation coefficient)

Variety	Germinating capacity (% of seeds germinated)	Topographical germinating capacity (% of seeds germinated)	r
Victory Freezer	65	66	
(Pisum sativum quadratum)	54	57	
	61	60	
	71	71	
	74	75	
			+0.786
Újmajori korai Viktória	61	62	
(Pisum sativum pachylobum)	69	70	
	75	80	
	96	92	
			+0.948

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DIFFERENCES BETWEEN TEXAS (Tcms) AND USDA (Scms) TYPES OF CYTOPLASMIC MALE STERILITY

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Significant differences between the Texas and the USDA types of cytoplasmic male sterility were obtained at both Martonvásár and Szentlőrinc for ear height and number of leaves above the top ear and only at Szentlőrinc for grain yield. There was no significant difference between the two types for plant height either in Martonvásár or in Szentlőrinc. The two types of cytoplasm do not interact with a given genotype in the same manner. Texas male-sterile is distinctly different from USDA male-sterile. The environment had a visible effect on fertility restoration.

Introduction

It is well-known that the cytoplasm may affect male sterility in inbred lines of corn and their hybrid combinations. The application of cytoplasmic male-sterility has become of increasing importance in corn breeding programs since about 1950. This may be due to the fact that detasseling process in hybrid seed corn production may be critical when unfavourable weather occurs, or when there is uncertainty of adequate labour.

In corn cytoplasmic male-sterility was first discovered by Rhoades (1931, 1933) in a Peruvian strain in 1931. Jones, starting in 1944, established a cytoplasmic basis for the pollen sterility of two plants in a progeny grown by M. T. Jenkins (Jones *et al.* 1957). This progeny traced back to a genetic tester stock (iojap × teopod). Jones and coworkers found that this source of cytoplasmic male-sterility was affected in its expression by the genotype. This source was named USDA or S type. In course of developing inbred lines from the variety Golden June, Rogers in 1944, noticed a male-sterile plant in one of the partially inbred lines. He outcrossed this plant, and found it to be another source of cytoplasm which induced male-sterility (Rogers–Edwardson 1952). This was the Texas or the T. type. It was found, too, to be affected by the genotype. Hadjikov (1959) presented the Moldavian type of cytoplasmic male sterility from a Moldavian variety.

In addition, there are in the literature descriptions of ten other discoveries of cytoplasmic male-sterility (Rhoades 1950, Jones *et al.* 1957, Briggle 1957, Alimova 1962). Jones (1951) found that plants from these various sources differed appreciably in maintenance of sterility when crossed with the same inbred lines. **Jones** (1954) and **Josephson** (1955) found that the USDA and Texas cytoplasms although superficially similar, were different in several ways. The most easily recognized difference was that a given genotype did not necessarily have the same degree of male-sterility in Texas cytoplasm as it had in USDA cytoplasm. **Li** *et al.* (1963) suggested that the M-type (Moldavian) cytoplasm might be similar to that of USDA-type.

Buchert (1961) discovered a fundamental difference between the USDA and the Texas cytoplasms. His conclusion was that in USDA cytoplasm only those microspores which contain the dominant gene for fertility restoration in USDA cytoplasm (Rf_3) will develop into fertile pollen grains, the microspores containing the recessive allele (rf_3) abort. In Texas cytoplasm, in contrast, all (95—100 per cent) pollen grains in a plant are fertile if the plant is at least heterozygous for the dominant gene or genes necessary for fertility restoration in Texas cytoplasm. **Buchert**'s basic conclusions have been confirmed by studies of **Duvick** (1965) with different materials.

In addition to the aforementioned differences, **Duvick** (1965) summarized the phenotypic differences between anthers of *T* and *S* sterile and partially fertile plants. Completely sterile plants in Texas cytoplasm characteristically do not exert anthers. In contrast to this many genotypes which are completely sterile in USDA cytoplasm will exert some or most of the anthers in the tassel. In partially fertile plants with Texas cytoplasm, anthers which have from about 10 to 95 per cent aborted pollen grains will usually exert, but will have a characteristic twisted, misshapen appearance. In plants with USDA cytoplasm which have from 50 to 95 per cent aborted pollen grains, the anthers are usually exerted also, but they tend not to be twisted and misshapen; rather they are of uniform dimension throughout their length.

Only limited information is available on the effects of Texas and USDA types of cytoplasmic male-sterility on hybrid performance. The present study was, therefore, undertaken as an attempt to investigate the effect of Texas type (Tcms) and USDA type (Scms) of cytoplasmic male-sterility on yields and other agronomic characteristics of some single and 3-way crosses.

Material and Method

The following terminology is used throughout this investigation:

T: The cytoplasm discovered by **Rogers** (1944); S: The USDA type of cytoplasmic male-sterility established by **Jones** (1944); M: The Moldavian type of cytoplasmic male-sterility presented by **Hadjinov** (1954); Cms: A cytoplasm that is associated with male-sterility in certain genic background.

To investigate the differences between Texas (Tcms) and USDA (Scms) types of cytoplasmic male-sterility the inbred lines C53 and P39 each was prepared by the two sources by back-crossing for n + 6 generations and were received from L. **Daniel** (Budapest).

In 1966 the four forms of the two inbreds were crossed as seed parents with mixed pollen from the following pollen parents, C5, M14, (M14 × 014), 014, Ia153, and A344, resulting in 24 single and 3-way crosses.

Crosses were arranged in a Troyan Square design with six replications (DARBY—GILBERT 1958).

This material was grown only in 1967 in two locations, Martonvásár on May 5 and Szentlőrinc on April 28. Each plot consisted of two rows each six meter long. Each row was planted by 15 hills and thinned to one plant per hill resulting in 30 plants per pot. In Martonvásár the land used has been cultivated only by corn since 1962. In autumn, 1965, form yard manure at a rate of 521 q/ha was added to the land before plowing. Pétisó containing 20.5 per cent $(NH_4)\ NO_3$ at the rate of 174 kg/ha was added in spring, 1966. Superphosphate containing 17.5 per cent P_2O_5 at a rate of 347 kg/ha, kálisó containing 40 per cent K_2O at a rate of 139 kg/ha were added in autumn, 1966. Linzisó containing 22.5 per cent $(NH_4)\ NO_3$ at a rate of 347 kg/ha was added in spring, 1967. After planting Hungazin at a rate of 4.3 kg/ha was added for weed control.

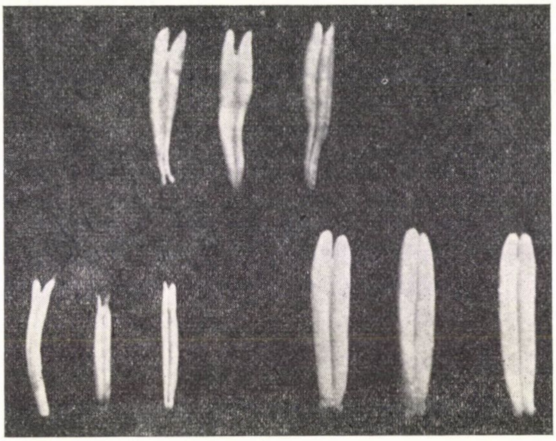

Fig. 1. Fertile, sterile and partially fertile anthers

In Szentlőrinc the land used was cultivated in 1966 by wild heliotrope (*Phacelia tancetifolia* Benth.). Farm yard manure at a rate of 347 q/ha was added to the land before plowing in 1966. Linzisó containing 22.5 per cent $(NH_4)\ NO_3$ at a rate of 347 kg/ha was added in spring, 1967. Before planting herbicides K-64 at the rate of 6.9 kg/ha, Afalon at a rate of 3.6 kg/ha and Arezin at a rate of 2.6 kg/ha were used for weed control.

Data were collected on 13 agronomic characters, among them the following: 1. Grain yield, 2. Plant height, 3. Ear height, 4. Number of leaves above the top ear, 5. Fertility restoration.

Grain yield data were obtained in kg per plot and converted to quintals (100 kg) per ha at 15 per cent moisture. Adjustments for yield were calculated in a few instances, on the basis of stand count when the plant population differed from 30 plants per plot.

To determine plant height and ear height 10 alternate plants per plot were measured from ground level to the first branch of the tassel and to the ear node. Measurements were recorded in cm.

The number of leaves above the top ear-node were counted on ten alternate plants per plot.

All progenies were examined for degree of sterility and the plants were classified as fertile, partially sterile, and sterile. Illustrations of the three classes are shown in Fig. 1.

All data were statistically analyzed to determine the significance of the difference between the mean values of the treatments according to the factorial mating design II. of COMSTOCK—ROBINSON (1948, 1952) where each of a group of parents used maternally are mated to each of another group of parents used paternally. All significant differences were calculated from the experimental error of each experiment.

The meteorological data during 1967 in Martonvásár and Szentlőrinc are presented in Table 1.

Table 1

Meteorological
Martonvásár, 1967 (Mv) and

	April		May	
	Mv	Szl	Mv	Szl
Number of rainy days	10	11	10	13
Rainfall, mm				
in the 1st ten days	20	24	15	10
in the 2nd ten days	10	28	10	114
in the 3rd ten days	30	34	60	27
Month-total	60	86	85	151
Mean of the years 1901—40	46	61	66	74
Deviation from the many years' mean	14	25	19	77
Daily mean temperature				
in the 1st ten days	10.1	10.2	16.0	15.9
in the 2nd ten days	14.1	14.8	19.5	19.2
in the 3rd ten days	10.3	10.4	17.2	18.5
Mean temperature in the month	11.5	11.9	17.6	17.9
Mean of the years 1911—50	10.1	10.6	15.9	15.6
Deviation	1.4	1.3	1.7	2.3

Results

Grain yield. Data presented in Table 2 show the effects of type of cytoplasm, pollen parents, and their interaction at Martonvásár and Szentlőrinc in 1967. Scms type of cytoplasmic male-sterility raised significantly the grain yield at Szentlőrinc. The effect of the pollen parents was significant at the two locations whereas, the interaction was only significant at Martonvásár.

Plant height. While there were no significant differences in plant height due to different types of cytoplasm the effects of pollen parents and the interaction between type of cytoplasm and pollen parents were significant at the two locations (Table 3).

Ear height. The effect of type of cytoplasm, pollen parents and their interaction on ear height are shown in Table 4. Scms type of cytoplasmic male-sterility gave significantly higher ears at the two locations of experimentation. In both Martonvásár and Szentlőrinc, the effects of pollen parents and the interaction of type of cytoplasm and pollen parents were significant.

Number of leaves above the top ear. Number of leaves above the top ear was significantly affected by type of cytoplasm and pollen parents. The interaction between type of cytoplasm and pollen parents had no significant effect (Table 5).

data
Szentlőrinc, 1967 (Szl)

June		July		August		September		October	
Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl
8	10	8	7	5	4	12	9	5	4
34	39	51	49	1	2	16	17	9	3
17	14	11	5	24	1	64	45	0	1
3	19	0	1	0	2	3	22	7	12
54	72	62	55	25	5	89	64	16	16
62	83	50	61	52	58	52	61	53	72
−8	−11	12	−6	−27	−53	37	3	−37	−56
18.5	18.8	22.9	23.2	23.9	25.1	21.0	22.5	15.0	16.9
17.1	17.6	24.4	24.3	20.9	21.7	16.3	17.6	13.1	17.2
24.7	23.2	25.0	27.4	19.5	22.3	17.0	19.6	11.2	13.7
20.1	19.9	24.1	25.0	21.4	23.0	18.1	19.9	12.2	15.9
19.1	19.2	21.5	21.3	20.7	20.8	15.7	16.2	10.6	10.8
1.0	0.7	2.6	3.7	0.7	2.2	2.4	3.7	2.1	5.1

Tcms type of cytoplasmic male-sterility reduced significantly the number of leaves above the top ear at both locations.

Fertility restoration. Data presented in Table 6 show number of total plants examined for fertility, total number of sterile, partially fertile, fertile plants, and the percentage of fertile plants at both Martonvásár and Szentlőrinc in 1967. In most cases fertility percentage was a little bit higher at Martonvásár than at Szentlőrinc. C5 as pollen parent had no ability to restore Tcms or Scms. Ole, Ia 153 and A344 restored completely Tcms and they had no ability to restore Scms type of Cytoplasmic male sterility.

Tcms type was restored by different pollen parents in a per cent of 59.3 and 57.8 at Martonvásár and Szentlőrinc respectively. The restoration percentage for Scms was 0.5 and 0.4 at both Martonvásár and Szentlőrinc respectively.

Table 2

Grain yield on 15% moisture in q/ha as influenced by type of cytoplasm, different pollen parents and their interaction

Martonvásár—Szentlőrinc, 1967

Type of cytoplasm	Pollen parent													
	C5		M14		(M14×O14)		O14		Ia153		A344		Mean	
	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl
Tcms	66.10	69.74	59.06	69.38	68.12	70.83	63.85	64.94	63.38	64.69	60.52	64.74	63.50	67.39
Scms	68.28	71.98	56.25	76.98	62.56	70.17	69.84	74.58	63.23	69.32	56.82	64.58	62.83	71.28
Mean	67.19	70.86	57.66	73.18	65.34	70.50	66.84	69.76	63.30	67.00	58.67	64.66	63.16	69.34

	Mv	Szl
L.S.D. for type of cytoplasm 1% level	NS	3,75 q/ha
L.S.D. for pollen parents 5% level	3,54	5,00 q/ha
L.S.D. for interaction 5% level	5,00	NS

Table 3

Mean height of corn plants in cm as influenced by type of cytoplasm, different pollen parents and their interaction

Martonvásár—Szentlőrinc, 1967

Type of cytoplasm	Pollen parent													
	C5		M14		(M14×O14)		O14		Ia153		A344		Mean	
	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl
Tcms	231.0	247.0	204.7	229.8	202.6	224.6	192.2	202.9	214.8	222.5	199.8	207.8	207.5	222.4
Scms	231.5	250.3	200.7	223.1	198.2	220.3	202.2	224.7	202.5	217.4	201.4	209.2	206.1	224.2
Mean	231.2	248.6	202.7	226.5	200.4	222.4	197.2	213.8	208.7	220.0	200.6	208.5	206.8	223.3

	Mv	Szl
L.S.D. for type of cytoplasm 1% level	NS	NS
L.S.D. for pollen parents 1% level	6,3	4,6 cm
L.S.D. for interaction 1% level	8,9	8,6 cm

Table 4

Mean height of ears cm as influenced by type of cytoplasm, different pollen parents and their interaction

Martonvásár—Szentlőrinc, 1967

Type of cytoplasm	Pollen parent													
	C5		M14		(M14×O14)		O14		Ia153		A344		Mean	
	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl
Tcms	83.4	78.6	71.5	75.1	68.1	71.8	58.5	58.0	71.4	72.8	67.9	68.2	70.2	70.8
Scms	81.9	82.2	73.2	76.5	69.2	72.4	68.0	72.2	72.3	72.2	69.3	68.1	72.3	73.9
Mean	82.6	80.4	72.4	75.8	68.6	72.1	63.3	65.1	71.9	72.5	68.6	68.1	71.2	72.4

	Mv	Szl
L.S.D. for type of cytoplasm 5% level	1,7	1,7 cm
L.S.D. for pollen parents 1% level	3,9	3,8 cm
L.S.D. for interaction 1% level	5,5	5,4 cm

Table 5

Mean number of leaves above the top ear as influenced by type of cytoplasm, different pollen parents and their interaction

Martonvásár—Szentlőrinc, 1967

Type of cytoplasm	Pollen parent													
	C5		M14		(M14×O14)		O14		Ia153		A344		Mean	
	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl
Tcms	5.45	5.56	5.80	5.83	5.24	5.33	4.75	4.52	5.36	5.39	5.09	5.08	5.28	5.29
Scms	5.68	5.71	6.00	5.80	5.44	5.48	5.00	4.84	5.34	5.47	5.21	5.05	5.44	5.39
Mean	5.57	5.63	5.90	5.82	5.34	5.41	4.88	4.68	5.35	5.43	5.15	5.07	5.36	5.34

	Mv	Szl
L.S.D. for type of cytoplasm 5% level	0,10	0,09
L.S.D. for pollen parents 1% level	0,24	0,22
L.S.D. for interaction 1% level	NS	NS

Table 6

Fertility restoration as influenced by different types of cytoplasm and different pollen parents
Martonvásár—Szentlőrinc, 1967

Pedigree	No. of plants				Pollen production				% of fertility	
	Total		Sterile		Partial		Fertile			
	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl	Mv	Szl
C53 Tcms × C5	178	184	178	179	—	3	—	2	0.0	1.1
,, × M14	180	176	127	141	39	26	14	9	7.8	5.1
,, × (M14×O14)	180	190	56	59	17	23	107	108	59.4	56.8
,, × O14	180	179	1	—	—	—	179	179	99.4	100.0
,, × Xa153	179	176	4	8	—	—	175	168	97.8	95.8
,, × A344	180	178	—	8	11	—	169	170	93.9	95.5
C53 Scms × C5	178	178	176	178	—	—	2	—	1.1	0.0
,, × M14	180	179	180	179	—	—	—	—	0.0	0.0
,, × (M14×O14)	180	179	180	178	—	—	—	—	0.0	0.0
,, × O14	180	184	180	184	—	—	—	—	0.0	0.0
,, × Ia153	180	175	179	175	—	—	—	—	0.0	0.0
,, × A344	180	172	178	172	2	—	—	—	0.0	0.0
P39 Tcms × C5	180	173	180	173	—	—	—	—	0.0	0.0
,, × M14	170	182	150	159	3	7	17	16	10.0	8.8
,, × (M14×O14)	180	177	50	31	27	51	103	95	57.2	53.7
,, × O14	180	188	8	11	—	—	172	177	95.6	94.1
,, × Ia153	180	173	—	14	10	—	170	159	94.4	91.1
,, × A344	180	178	12	15	—	—	168	163	93.3	91.6
P39 Scms × C5	180	181	171	181	9	—	—	—	0.0	0.0
,, × M14	180	131	177	130	3	1	—	—	0.0	0.0
,, × (M14×O14)	180	153	170	145	6	4	4	4	2.2	2.6
,, × O14	178	135	162	131	13	—	3	4	1.7	3.0
,, × Ia153	179	143	178	143	1	—	—	—	0.0	0.0
,, × A344	178	178	174	178	4	—	—	—	0.0	0.0
Type of ctyoplasm										
Tcms	2147	2154	766	798	107	110	1274	1246	59.3	57.8
Scms	2153	1987	2105	1974	38	5	10	8	0.5	0.4

Discussion

Concerning the differences between Tcms and Scms male-sterility, until this investigation no data were available in the literature about the differences between the two types in yield and other agronomic characteristics.

Yield data presented in Table 3 show that crosses having Scms male-sterile surpassed those having Tcms type in the average of the two locations by 2.2 per cent. C5 as a pollen parent surpassed A 344 in the average of the to locations by 2.9 percent. It is interesting to see that the combinations of Scms × C5 surpassed those of Tcms × A 344 in the average of the two locations by 9.3 per cent.

It is an evidence that differences obtained in yield apparently were dependent upon the interaction of a specific genotype with a specific cytoplasm.

Besides the yielding ability, plant height, ear height, and number of leaves above the top ear were affected significantly by the interaction between type of cytoplasm and pollen parents.

It seems also that particular genotypes, cytoplasm, and their interactions appear to be important in expression of these characters.

Data presented in Table 6 show that the Texas male-sterility was distinctly different from USDA male-sterility. Testers that restored fertility to the Texas type did not act as restorers to the other male-sterile. A completely male-sterile progeny was produced when C53 Scms was pollinated by the six pollen parents. When P39 Scms was pollinated by the same pollen parents, it gave a varying number of partially fertile plants. The data in Table 6 show variability in degree of expression of fertility restoration among crosses between a common tester and different lines bearing the same source of sterility. Some male-sterile parents may have been heterozygous for a modifier gene or genes which, when complemented by the male parent genotype, resulted in sterility.

Most F_1 populations involving (M14 × 014) pollen parent segregated for sterility, suggesting that the single cross may have been heterozygous for the restorer gene or genes. M14 pollen parent interacted specifically with the Texas male-sterile giving F_1 populations predominant in sterile plants.

Modifiers in inbred lines influence the expression of cytoplasmic male-sterility to a greater degree in USDA than in the Texas male-sterile.

The environmental influence on fertility restoration can be easily detected from Table 6. In most cases percentage of fertility restoration was higher at Martonvásár than at Szentlőrinc. Meteorological data in Table 1 indicate that at the time of flowering (July) the total rainfall at Martonvásár was 62 mm and the mean temperature was 24.1 °C, whereas, in the same period at Szentlőrinc, the total rainfall was 55 mm and the mean temperature was 25 °C.

Li *et al.* (1963) suggested that (M-type) Moldavian cytoplasm might be similar to that of S-type. It was found that 014 as a tester restored completely the Moldavian sterile cytoplasm, also M14 in combination with the Moldavian sterile cytoplasm gave 50 per cent and 80.8 per cent fertile plants in 1966 and 1967 respectively (Oraby 1968). Nearly all F_1 populations resulted from Scms cytoplasm in combinations with 014 or M14 were sterile at both Martonvásár and Szentlőrinc.

This is a clear indication that (M-type) Moldavian cytoplasm is not similar to that of (S-type) USDA cytoplasm, or there are other genes in both C53 Scms and P39 Scms which masked the effect of the two testers 014 and M14 in restoring the (S-type) and those genes may not be found in the inbred line WF9 Mcms.

Acknowledgement

Here I would like to express my deep gratitude to S. Rajki, Director of the Agricultural Research Institute of the Hungarian Academy of Sciences, Martonvásár for his continuous help during the progress of the work. Heartily thanks are also due to J. O'svÁth from the Martonvásár Institute for his cooperation in designing the experiments and analyzing the results statistically. Heartily thanks are also due to L. Daniel from the Institute of Genetics, Budapest for supplying the initial stock for this investigation.

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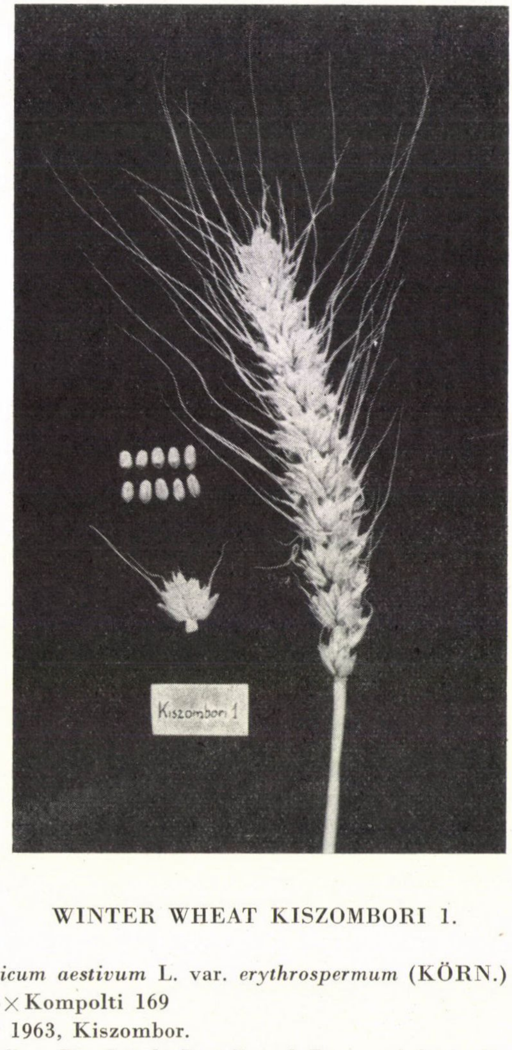
VARIA



WINTER WHEAT KISZOMBORI 1.

Taxonomic place: *Triticum aestivum* L. var. *erythrospermum* (KÖRN.) MSF.
Origin: Produttore 13×Kompolti 169
Beginning of breeding: 1963, Kiszombor.
Breeder: Dr. János Lelley, Dr. László Parádi and Zsigmond Sarkadi.
State qualification: Provisionally certified improved variety, 1968.
General characterization: Fairly winter-hardy and drought-resistant medium early variety, with aristate ears and red grains, good productivity and medium flour quality. Its yield level is close to that of Bezostaya 1.
Morphological description:
  Root system: roots penetrate the soil to nearly 1 m; root system rather strong.
  Shoot system: vigorous development, favourably productive stooling, high stability (4.5) and an extreme capacity for regeneration (SZABÓ 1969).

Straw: medium high, its height is an average of 88 cm (ranges from 85 to 92 cm), thick, strong, light yellow.

Foliage: generally upstanding (even when young), linear lanciform, broad, ash-green, arched leaf blade.

Ear: awned, fusiform, whitish-yellow; mature ears are drooping. Average number of flowers in the spikelets is 3, readily pollinated; number of grain per ear: 19—22. Number of ears per m^2: 460—470. Weight of ear: 0.804 g. Lamellae of short awned glumes are oval, apices simple.

Caryopsis: squat egg-shaped with a blunt apex; 6 mm long and 3.5 mm broad; rounded square scutellum. Red-brown grain, with slightly glassy fracture. Intensive ventral line. Thousand-grain-weight 41—44 g. Medium flour quality (B_1—B_2). Not inclined to dropping though easy to thrash.

Biological characters:

Germination: minimum temperature +2, maximum 35°C.

Vegetative period: from sowing to maturity an average of 260 days.

Development: rapid and vigorous, early earing. Development and ripening somewhat quicker than in Bezostaya 1.

Winter hardiness: good, though lower than in Bezostaya 1.

Disease resistance: while somewhat susceptible to leaf- and stem rust, still escapes infection, due to its rapid development.

Farm technology requirements:

Sowing: according to the breeder's suggestion it is to be sown early, possibly at the beginning of October. Sowing depth is 5—6 cm in a settled soil. Seed requirement is 3.4—3.6 million germs/cad.yoke (1 cad.yoke = 5754.56 m^2).

Soil requirement: warm soils with good water management and rich nutrient content. It makes high yield returns for large fertilizer doses (SZABÓ 1969).

Productivity: 21—26 q/cad.yoke (= 5754.56 m^2).

Area of cultivation: successfully grown in the middle and southern regions of Hungary; its production is recommended for irrigated areas.

*

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GENETIC INVESTIGATION IN AEGILOPS

Investigations into the species-relationship of the genus *Aegilops* dates back to the early part of the present century. Starting from the pioneer work of Kihara and his associates this aspect has been dealt with by different workers all over the world. The genome-analytical work of the Kihara school ultimately led to the formulation of specific genome symbols for almost all the species of *Aegilops* (KIHARA 1963). However, in spite of this thoroughness of Kihara's monumental work, the different species of *Aegilops* do not always behave in accordance with their genome symbols. Perhaps, this is because of the fact that in certain cases the symbols were not assigned to the different species on the basis of direct and concrete genome analytical work but with the help of indirect evidence (CHENNAVEERAIAN 1960). We feel that in certain cases the present genome-symbols of *Aegilops* species need some sort of modification to be brought into agreement with practical experience. The purpose of the present investigation was to study the affinity among different species of *Aegilops* through hybridization.

Table 1

Record of hybridization between Aegilops species (diploid × diploid)

Combinations	Year	No. of flowers crossed	No. of seeds obtained	Percentage of seed-setting	Percentage of sprouting
Ae. speltoides ×	1968	236	0	0	0
Ae. longissima	1970	120	0	0	0
Ae. umbellulata × *Ae. squarrosa*	1969	30	0	0	0
Ae. umbellulata × *Ae. mutica*	1969	24	1	4.17	0
Ae. umbellulata × *Ae. caudata*	1969	24	0	0	0
Ae. mutica × *Ae. sharonensis*	1969	24	0	0	0
Ae. mutica × *Ae. speltoides*	1969	28	0	0	0
Ae. squarrosa × *Ae. mutica*	1969	162	0	0	0
Ae. caudata × *Ae. squarrosa*	1970	80	0	0	0
Ae. caudata × *Ae. speltoides*	1969	100	0	0	0
Ae. caudata × *Ae. squarrosa*	1970	54	0	0	0
Ae. speltoides × *Ae. caudata*	1970	220	0	0	0
Ae. caudata × *Ae. speltoides*	1970	120	0	0	0
		1270	1	0.08	0

Experiments were conducted at the experimental garden of the Agricultural Research Institute, Martonvásár during 1968—70. Plants were raised with a spacing of 30×20 cm. Each year crosses were made between all the possible combinations of *Aegilops* species. The emasculation was done with the help of a fine forceps and pollination was done 2 days after emasculation by placing 2 mature anthers in each flower. Wherever possible, reciprocal crosses were made to facilitate the process of seed-setting.

Hybrid seeds obtained from the succesful crosses were sown in the next season to study the extent of sprouting. After sprouting, critical observations were made regarding hybrid lethality and survival. After harvest the seed-yield of hybrid F_1 plants was determined in the laboratory.

Crossability. Close observation of Table 1 shows poor compatibility among the diploid species of *Aegilops*. Out of a total 1270 flowers crossed, only 1 seed was obtained. This single succesful seed-setting was observed in *Ae. umbellulata* × *Ae. mutica*, however, even this single seed was infertile. It is to be admitted in this connection that the number of flowers crossed in certain combinations was not adequate.

In the second group of interspecific hybridization, that is between diploid and tetraploid plants there was slight improvement in seed-setting as eight seeds were obtained out of 448 flowers crossed (Table 2). In the hybridization between *Ae. ovata* and *Ae. longissima*, a few seeds were obtained when the tetraploid plant was used as mother. However, this cannot be accepted as the general trend, as in other cases where a tetraploid *Aegilops* acted as the female parent, no seed was obtained, whereas diploid o× tetraploid o did produce seeds as evidenced by the *Ae. squarrosa* × *Ae. crassa* and *Ae. squarrosa* × *Ae. ventricosa* crosses.

Table 2

Records of hybridization between Aegilops species (diploid × tetraploid)

Combinations	Year	No. of flowers crossed	No. of seeds obtained	Percentage of seed-setting	Percentage of sprouting
Ae. longissima × *Ae. cylindrica*	1968	90	0	0	0
Ae. ovata × *Ae. longissima*	1968–69	24	4	16.67	75.00
Ae. squarrosa × *Ae. ovata*	1969	48	0	0	0
	1970	112	1	0.90	0
Ae. ovata × *Ae. squarrosa*	1970	54	0	0	0
Ae. triuncialis × *Ae. caudata*	1969	42	0	0	0
Ae. squarrosa × *Ae. crassa*	1969	34	1	2.94	0
Ae. squarrosa × *Ae. ventricosa*	1969	44	2	4.76	0
		448	8	1.78	

The situation is slightly improved in the case of tetraploid × tetraploid crossings, perhaps because of the uniformity in chromosome number of the two parents. The highest percentage of seed-setting was 8.33, obtained from *Ae. triuncialis* × *Ae. biuncialis*, and its reciprocal. Combinations of *Ae. triuncialis* and *Ae. biuncialis* with *Ae. ovata* were unsuccessful when *Ae. ovata* was used as the male parent. On the other hand, crosses between *Ae. ovata* and *Ae. biuncialis* having *Ae. ovata* as the female parent gave a seed yield of 4.17 per cent (Table 3).

With the exception of three instances, hybrid seeds were infertile in all cases. In the case of *Ae. ovata* × *Ae. longissima* and *Ae. triuncialis* × *Ae. biuncialis*, the percentages of sprouting were 75.00 and 100.00 respectively. The sprouting was also quite satisfactory in *Ae. biuncialis* × *Ae. triuncialis* (Table 3).

Examination of fertility. We could not make any pollen fertility examinations in the three interspecific F_1 combinations. However, all the three hybrid combinations produced seeds in the F_1 generation. The seeds obtained from *Ae. ovata* × *Ae. longissima* were all shrivelled and did not germinate in the corresponding generation. The other two combinations gave surprisingly high yield. Table 4 gives an account of the fertility of these hybrids.

Our observations show very poor compatibility within the genus *Aegilops*. The diploid × diploid interspecific crosses set seed in only one instance. No hybrid seed was obtained even in those cases where the two parents came from the same genomic group, for example, *Ae. speltoides* × *Ae. longissima* and *Ae. umbellulata* × *Ae. caudata*. In connection with the failure of seed-setting in the latter case, it can be said that the genomes of umbellulata (C^u) and caudata (C) need further intensive examination for the determination of their exact relationship. Kihara's nomenclature was doubted by SEARS (1948) as an overemphasization of the closeness of relationship.

Table 3

Records of hybridization between Aegilops species (tetraploid × tetraploid)

Combinations	Year	No. of flowers crossed	No. of seeds obtained	Percentage of seed-setting	Percentage of sprouting
Ae. triuncialis × *Ae. ovata*	1969	28	0	0	0
Ae. ovata × *Ae. biuncialis*	1969	48	2	4.17	0
Ae. biuncialis × *Ae. ovata*	1970	24	0	0	0
Ae. cylindrica × *Ae. triuncialis*	1970	60	1	1.67	0
Ae. triuncialis × *Ae. biuncialis*	1969	24	2	8.33	100.0
Ae. biuncialis × *Ae. triuncialis*	1969	84	7	8.33	28.57
		268	12	4.48	

Table 4

Seed production in F_1 hybrids

Combinations	Year	No. of plants examined	Seed/plant	Seed/ear
Ae. ovata × *Ae. longissima*	1968—69	2	8.0	1.78
Ae. biuncialis × *Ae. triuncialis*	1970	2	115.5	2.16
Ae. triuncialis × *Ae. biuncialis*	1970	2	266.0	4.88

Among the diploid × tetraploid combinations, seed-setting in *Ae. squarrosa* × *Ae. crassa* and *Ae. squarrosa* × *Ae. ventricosa* is facilitated by their possessing one common genome (D). The seed-setting observed in the case of *Ae. triuncialis* (CC^u) and its reciprocal was quite high and the fact of their having a common genome does not fully explain this. The sprouting of hybrid seeds and the fertility of hybrid plants in the F_1 generation were remarkably high in this combination in both directions. In fact, the seed-production in the F_1 generation was so high that it gave the impression of the existence of some closer relationship between these two species. We could not make cytological observations in these two combinations. CHENNAVEERAIAH (1960) in his karyomorphologic studies in *Ae. biuncialis* and *Ae. triuncialis* expressed doubt about the identity of their second genome (M^b and C respectively). He pointed out that the genome-type C^uM^b designated by the Kihara school was not based on any direct genome-analytical work but probably on the morphological characters and karyotype study made by previous workers (SENYANINOVA-KORCHAGINA 1930). On the basis of his own study, he claimed that the M^b genome of *Ae. biuncialis* might have come from a so far unknown source. *Ae. triuncialis* makes a highly variable species complex. On the basis of their success in synthesizing this species from *Ae. caudata* (c) and *Ae. umbellulata* (C^u), KIHARA—KONDO (1943) attributed the CC^u genomic formula to the tetraploid *Ae. triuncialis*. However, the C genome is highly variable in this species complex. **Chennaveeraiah**, after analysing his karyotypic observations, concludes that in some taxa of *Ae. triuncialis*, either the C genome has undergone significant modification or some foreign genome is present instead of C genome. Our data on the crossability and fertility of hybrids between these two species strengthens this view.

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ROLE PLAYED BY PARTS OF FLOWER IN THE TRIPPING MECHANISM OF ALFALFA (MEDICAGO SATIVA L.) FLOWER

The flower of alfalfa does not open even when completely developed (Fig. 1). At this stage, however, the staminal column* snaps immediately as a result of the influence of external stimuli (Fig. 2), the membrane which protects the stigma becomes damaged, the pollens come into contact with the secreta of the stigma and pollination, and consequently fertilization becomes possible. According to the researchers' opinion the opening of the flower — which can be caused by insect visitation, machines constructed for this purpose as well as climatic conditions — is an indispensable precondition of pollination and fertilization.

The structure of the flower has been described by many authors; however, the role played by the individual parts of the flower in the tripping mechanism has been but superficially dealt with during the investigations, and the results published are contradictory (Heszky 1970).

On the basis of the results of our investigations it has been necessary to make a distinction between the closing and the opening apparatus in the structure of the flower.

The closing apparatus consists of those organs of the flower the connection of which keeps the staminal column in the keel and prevents it from bending.

Those parts of the flower which do not belong to the closing apparatus can be removed from the flower without the bending of the staminal column. With the preparation method evolved by Heszky (1970) the following flower parts can be removed without the opening of the flower: 1. calyx (Fig. 3), 2. standard (Fig. 3), 3. the blades of wings (Fig. 4), 4. the hornlike projections of wings (Fig. 5), 5. the small projections of wings (Fig. 6).

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* Filament tube and ovary.

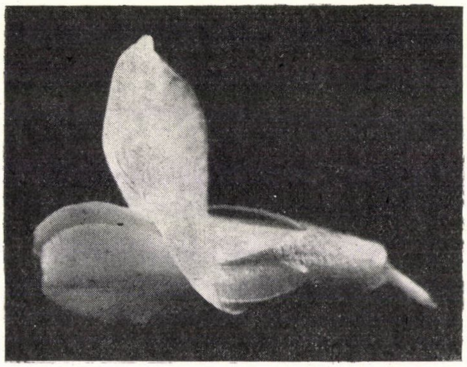

Fig. 1. Side view of the alfalfa flower

Fig. 2. Side view of the flower after tripping

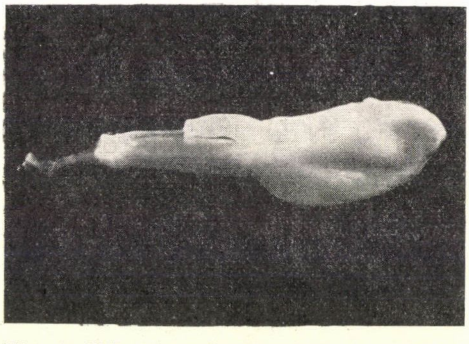

Fig. 3. Side view of a closed flower without calyx and standard

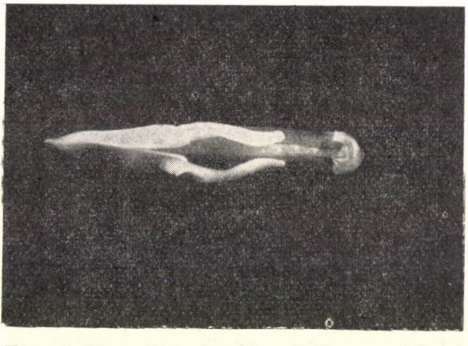

Fig. 4. Top-view of a closed flower without calyx, standard and blades of wings

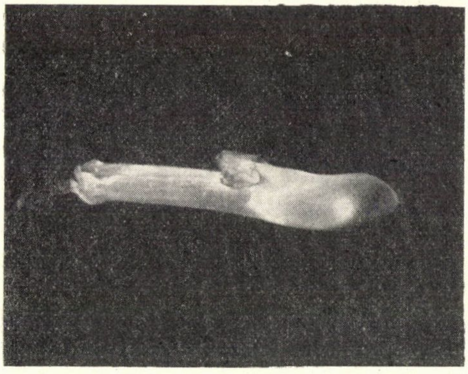

Fig. 5. Side view of a closed flower without calyx, standard, blades of wings and horn-like projections of wings

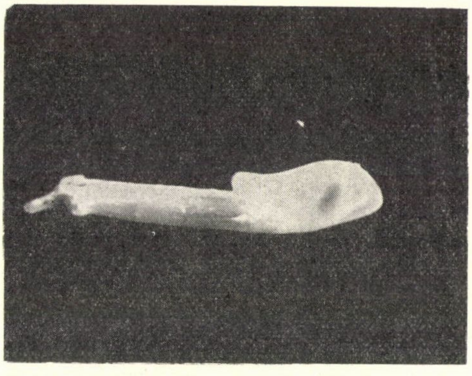

Fig. 6. Side view of a closed flower without calyx, standard, blades of wings, horn-like projections and small projections of wings

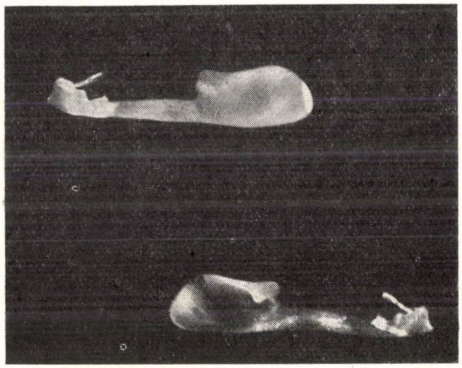

Fig. 7. Components of the closing apparatus of the flower; above: from outside the flower, below: from inside the flower

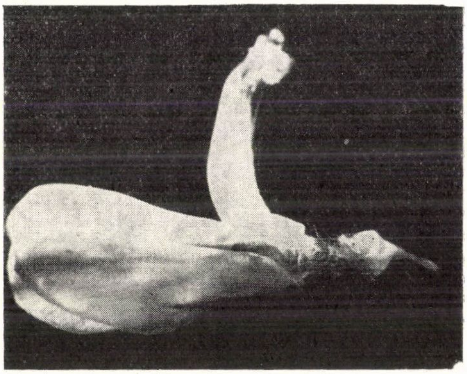

Fig. 8. Side view of the bending form of the staminal column after keel blades have been separated

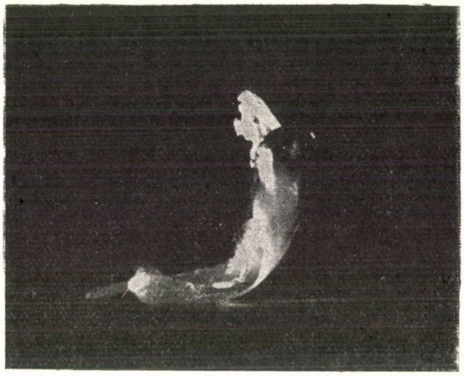

Fig. 9. Side view of the bending form of the staminal column after the upper edges of the keel blades have been separated

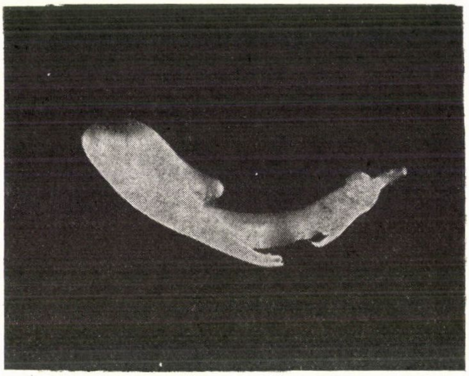

Fig. 10. Side view of the bending form of the staminal column after the claw of the keel has been cut through

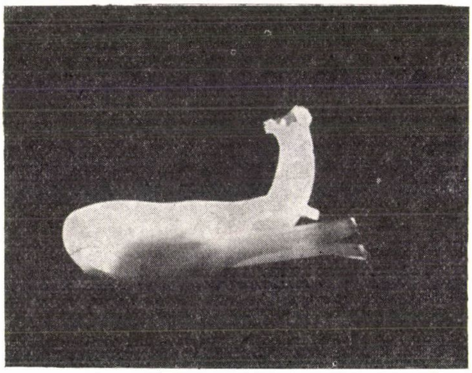

Fig. 11. Side view of the bending of the staminal column after the receptacle has been removed

Fig. 6 shows that the staminal column remains in a state of tension between the blades of the keel, the claw of the keel and the receptacle.

Thus, the staminal column is closed by these three organs (Fig. 7). Any damage done to, or the removal of either the receptacle, or the claw of the keel or the blades of the keel resulted in every case in the immediate intensive bending of the staminal column. However, the way the staminal column bent depended on the organ affected.

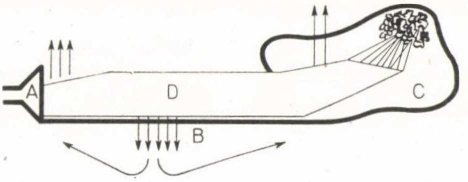

Fig. 12. Operation of the closing apparatus: A: receptacle, B: claw of the keel, C: blades of the keel, D: staminal column

1. When the blades of the keel were laterally pulled apart tripping set in at once (Fig. 8).
2. Separating the upper part of the keel blades and moving the blades toward the basal part of the staminal column, also resulted in the column being bent completely. At the upper, open part of the keel there appeared the anthers and the stigma. However, at other parts of the keel the blades did not separate (Fig. 9).
3. When the claw of the keel was cut through with a scalpel, the staminal column bent again without the keel opening (Fig. 10).
4. Removal of the receptacle resulted in the staminal column bending without the keel opening (Fig. 11).

The four types of beding well illustrate the importance of the individual components in the closing apparatus. The receptacle is the common base for fixing the claw of the keel and the staminal column, the latter being thus prevented from bending upwards (Fig. 12). The highest resistance to the tension of the column — which wants to bend at both ends — is displayed by the claw of the keel (Fig. 12). The pressure of the column is the lowest at the blades of the keel, therefore even the tooth-like projections described by LARKIN—GRAUMANN (1954) are able to prevent the column from snapping up.

The opening apparatus consists of those organs of the flower through the connection of which wild bees are able to force open the coalescent sepals of the keel.

It follows from the above that in situ opening of the flower is only possible when the connection between the coalescent blades of the keel has become loose. Thus the flower can be opened only by the — indirect or direct — crosswise removal of the keel blades.

Fig. 7 shows two cavities on the blades of the keel: one on the upper surface, the other on that facing tha basal part of the flower. It is into these cavities that the small projections of the wings — which originate from the protrusions of the blades of wings — reach (Fig. 13). The small projections of the wings are not grown together with the cavities of the keel; they can be readily removed without the opening of the keel (Fig. 6).

Through this connection any displacement of the wings results in the displacement of the keel blades. In the case of insect visitation the blades of the wings move downward, and through the small projections and the folds of the keel the motion is transferred to the blades of the keel (Fig. 14), which move away and the staminal column snaps forward.

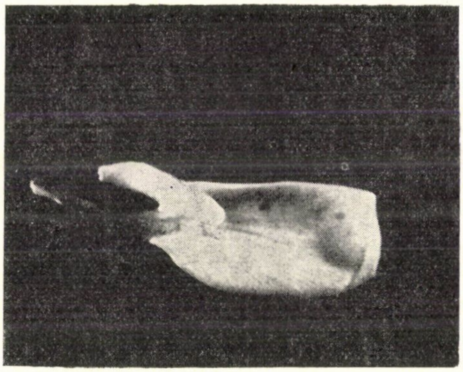

Fig. 13. Wing, from the inside of the flower

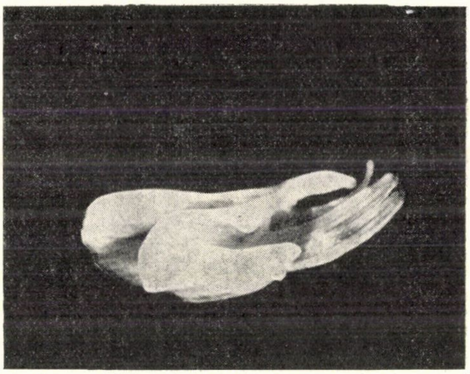

Fig. 14. Opening apparatus. Connection of wings and keel is visible

The floral parts forming the closing apparatus of the flower are: receptacle, claw of the keel, blades of the keel.

The floral parts forming the opening apparatus of the flower are: cavities of the keel blades, small projections of wings, blades of wings.

*

Prepared at the National Institute of Agrobotany, Tápiószele

L. Heszky

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RETARDING AND PROMOTING EFFECTS OF N-DIMETHYL-N-(β-CHLOROETHYL) HYDRAZONIUM CHLORIDE (CMH) ON WHEAT SEEDLINGS*

In a previous work it was reported that the hydrazonium compounds showed growth retarding effects on wheat and other plants (Jung 1967a, b). Among the compounds tested are: N-Dimethyl-N-(β-chloroethyl) hydrazonium chloride (CMH), N-Dimethyl-N-(β-bromoethyl) hydrazonium bromide (BMH), N-Dimethyl-N-isopropyl hydrazonium bromide (IMH) and N-Dimethyl N-allyl-hydrazonium chloride (AMH). The first one, namely CMH was studied the most intensively. Using this compound under Egyptian conditions as spray to the wheat variety G.150 (*Triticum vulgare*), low concentrations led to stimulation in plant height (El-

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* In the first experiment all concentrations of the CMH-preparation retarded consequently the growth processes of wheat varieties, while in the second experiment authors registered a stimulative effect. Considering the great interest shown in the question of growth inhibitors we publish Jung et al.'s study in spite of its disputable results. (Editorial office.)

Fouly *et al.* 1970). This effect was apparent some days after treatment till maturity. Only high concentrations led to a slight reduction in stem height.

As this result is not in accordance with those of **Jung** (1967a, b) the following study was conducted to verify the physiological effect of CMH on the height of wheat seedlings.

Two varieties of wheat were used: Opal, a German one, which showed retarding effect due to CMH and G.155 an Egyptian one, which showed a stimulatory effect.

Two experiments were conducted in both Germany and Egypt under the environmental conditions of each country.

The first experiment was carried out in Germany during March, 1969. Seeds of the two varieties were sown in Neubauer pots (100 seeds/pot), each filled with 600 g soil. CMH was used as either soil drench or as foliar spray in different doses. Applying CMH as soil drench was carried out by watering each pot with 20 ml of the solution containing the given dose per pot before sowing. The foliar spray was performed 6—7 days after sowing, when plants were about 6—8 cm in height. Each pot received 100 mg N during the growth period. Each treatment was replicated in 3 pots and the whole experiment was repeated two times.

The second experiment was carried out in Egypt during March, 1970 under the same conditions mentioned above, using the same experimental procedure.

A third experiment was conducted in Egypt only, in which the soil treatment was carried out on the same day as the foliar application and followed by light irrigation. The aim of this experiment was to exclude the effect of the different lengths of the exposure periods to the growth regulator in both kinds of treatment.

In all experiments stem height was measured when the check plants were about 20—25 cm in height.

Table 1 exhibits the results of the two first experiments which were similarly carried out. The following can be detected from the table:

Table 1

Effect of CMH on height of wheat seedlings

CMH Concentration (mg/pot)	Height %			
	Soil application		Foliar spray	
	Opal	G. 155	Opal	G. 155
First experiment				
0	100.0	100.0	100.0	100.0
1.5	—	—	89.0	97.0
3.0	79.9	87.0	88.0	96.0
6.0	75.6	82.0	90.0	93.0
12.0	70.2	82.0	—	—
Second experiment				
0	100.0	100.0	100.0	100.0
1.5	96.5	102.5	102.8	113.1
3.0	94.4	110.9	108.1	106.4
6.0	89.6	98.0	109.7	113.1
12.0	84.8	95.0	101.2	116.5

1. Different varietal responses to CMH treatments in both experiments. The variety Opal always showed a stronger retarding effect due to CMH than the variety G.155. Moreover, spraying CMH in the second experiment resulted in a stimulatory effect upon the two varieties, which was more pronounced in G.155 than in Opal. Such varietal changes in the response of wheat to growth regulators are well known (LINSER 1968) and might be due to the physiological differences of the different varieties. This can be successfully used to study the mechanism, by which these chemicals affect plant growth.

Table 2

Effect of CMH used as soil drench at different growth stages on height of wheat seedlings

CMH Concentration (mg/pot)	Height %			
	Opal		G. 155	
	before sowing	7 days after sowing	before sowing	7 days after sowing
0	100.0	100.0	100.0	100.0
1.5	96.5	95.2	102.5	103.0
3	94.4	95.2	111.9	111.9
6	89.6	92.0	98.0	104.4
12	84.8	84.3	95.0	96.9

2. The mode of application seems to influence the intensity of the apparent effect on plant height in both varieties and in both experiments. Using the same concentration, CMH as soil drench always had a more retarding effect than the foliar spray. Moreover, in the second experiment soil application of CMH to Opal decreased height proportional to the increase of the CMH concentrations used, while the foliar application led to an increase in stem height up to a certain concentration, then it declined. In the first case this effect can be attributed to the duration of the exposure period of the plants to the CMH effect according to the different application times of the two methods.

In the third experiment, CMH was applied at the same time as soil drench or as foliar spray. Comparing the effect of soil drench applied before sowing or with the foliar application, 7 days after sowing (Table 2) no differences could be found. Therefore, the differences in the CMH effect due to the method of application cannot be due to the prolonged treatment period as it might have been concluded from the results of the first and second experiments. This effect might be correlated with the method of application itself, and may be explained by possible differences between absorption of CMH through roots and its penetration through leaves.

3. In the first experiment, which was performed under German conditions, both varieties showed a more or less retarding effect due to CMH treatment, regardless of the method of application. However, in the second experiment, which was conducted under Egyptian conditions, only the variety Opal showed a retardation in all CMH doses used, when applied to soil, while G.155 showed under the same treatment conditions a stimulation at the lower doses and a retardation, only when high doses were applied. On the other hand foliar spray under Egyptian conditions had a stimulatory effect on the height of both varieties. This effect was, however, less exhibited in the variety Opal and was not observed in plants receiving the highest CMH dose.

These results indicate that CMH effect on plant height depends on the interaction between the varietal response, method of application, concentration used and the environmental conditions. Other chemicals, which have growth regulating characters exhibit different effects due to environmental conditions, which influence plant metabolism. So, it was reported that CCC showed stimulating effects on height with different plants (Halevy—Wittwer 1965, Halevy—Shilo 1970). Moreover, differences in its effect due to mode of application, as well as day-length were also reported (Wünsche 1969). In addition, temperature modifies CCC action (Heide 1969, El-Fouly—Moubarek—unpublished). Also abscisic acid was found to exhibit growth promoting and inhibiting effects in spirodela (Van Staden—Borman 1969).

The presented results indicate that the effects of these growth regulating chemicals on metabolism under different environmental conditions leading to different morphological responses are different and seem to be a good tool towards understanding the mode of action, by which they induce changes in plant growth.

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THE OLDEST HUNGARIAN APIARIST BOOK[1]

Habent sua fata libelli — books have their own fate. Well, the fate of the oldest Hungarian apiarist book was that it should not to survive in its original form. Up to the present day only various copies have been found.

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[1] The title of the book: "*Liber Apiaster Opus tam Vtile, quam necessarium. Cum experimentiis rebusque probatis Nicolai Horhi Apiastri Principalis Waradiensis.* — Méhész könyvetske Meljben egészben leiratik a Méhek körül való dajkálkodásnak igaz módgya és hasznos mestersége, Melljet a Tekéntetes és Méltóságos Fejedelemnek, az öreg és iffiu Rakotzi Györgjnek Fő Méhész Mestere Horhi Miklós, mint rend szerint való hivatalát egész életének foljásában Nagj Váradon laktában tiszti szerint 37 esztendeig követett. (Apiarist booklet in which the method and useful profession of bee rearing is fully contained. It was practised by Miklós Horhi, leading apiarist of the Honourable Princes György Rákóczi senior and junior as a profession throughout his life who lived at Nagyvárad, for 37 years.) *Varadini anno Domini 1645. Excudebat Abrahamus Szentzi.* — See: Bibliographia Oeconomica Hungariae. Bp. 1934. No. 71.

Past of apiculture. Apiculture looks back upon a brilliant past in Hungary. For many centuries honey has been — here too — an indispensable element of nutrition, wax the noblest material used for lighting, and bragget an alcoholic drink as much-sought as among Germanic and Slavic people.[2] That is what the old proverb refers to: "Bees sweeten our days and light up our nights".[3] As early as in 1055 in a deed of foundation of King András I. (1046—1060) not only the apiaries but the apiarists are also mentioned.[4] In County Baranya for example royal apiarists existed who up to 1370 belonged to the Bodvölgy possessions.[5]

In the Middle Ages even serfs were occupied with apiculture in Hungary which is proved by the fact that in 1514, after the suppression of the peasants' rising led by György Dózsa to overthrow the feudal regime the Diet ordered the serfs to give tithes from their beehives to the landlords. This form of taxation was only cancelled by the Diet of 1836.[6]

The era in which the "Apiarist booklet" was written. After the Mohács Disaster in 1526 the position of Hungary was determined by the Turkish rule and the accompanying permanent war-like conditions over more than one and a half centuries. Hungary was divided into three parts. Transylvania became a principality which was reduced to a vasalage by the Ottoman Porte but remained independent in its internal affairs. Transylvania was called "Fairyland" by the contemporaries. After Gábor Bethlen (1580—1629), one of the greatest Hungarian historical characters of the 17th century, György Rákóczi I. (1593—1648) became Prince of Transylvania, under whose reign the principality developed in peace. These events were characterized by one of our literary historians in the following way: in the 16th century the division of the country was rather a "political necessity", while in the 17th century it was a "spiritual reality". Cultural life in the Hungarian kingdom and in Transylvania showed different trends.[7]

György Rákóczi I. and his wife Zsuzsanna Lórántffy (cca. 1600—1660) did much for the development of education and literature. It was in their house that Martin Opitz (1597—1639), the reorganizer of German poetry, was friendly received. It was to them that Comenius Ámos János (1592—1670), the great scientist who had been tossed about a good deal fled and was then teacher at Sárospatak for four years. It was here too that he wrote his pioneer hand-book on the demonstrative method of instruction: *Orbis Sensualium Pictus* (The visible world in pictures). János Apáczai Csere (1625—1659), the outstanding pioneer of Hungarian culture, education and science, when returning from Holland became teacher also on the estate of the Rákóczi-house, and it was there that he published his great works. It was again György Rákóczi I. who covered the expenses of publishing the books of Alstedius, Piscator and Bisterfeld.

All this has been told because Nicolaus Horhi, the author of the first Hungarian apiarist book was the leading apiarist in the Rákóczi house for 37 years. Of all the Hungarian libraries in the 17th century the Rákóczi Library was the richest in Hungarian prints.[8]

Description and evaluation of the apiarist booklet. As it was said at the beginning, this work is also one of those which were lost. Only various copies have remained. Two copies are among the manuscripts of the National Széchényi Library. One of them was prepared in 1720 and consists of eight quarto sheets. All the sheets are damaged in the middle, so the text is incomplete. Its call number is: Quart. Hung. 2327. The other copy was prepared in 1802; it extends to ten octavo sheets. This copy is intact. Its call number is: Oct. Hung. 1076.

In print it was published by Kálmán Kenessey in the journal "Kertész Gazda", 1866. No. 22. after a certified copy made by István Bogárdi Mészöly, judge of the Zala County Court,

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[2] Jenő Rodiczky dr.: A Méhről . . . (On the bee . . .). Magyar-Óvár 1876. 70—88. pp.

[3] Kálmán Sőtér: A méh és világa (The bee and its world). II. Bp. 1908. 69 p.

[4] *In apibus et ipsorum custodibus*. Fejér, Codex diplom. I. 388—389. pp.

[5] Századok 1877. 166 p. Gusztáv Hatos: Királyi méhészek Baranyában (Royal apiarists in County Baranya).

[6] VII. tc. 4. §. Cited by dr. Rodiczky: A Méhről . . . (On the bee . . .) pp. 71—72.

[7] Antal Szerb: Magyar irodalomtörténet (Hungarian literature history). Bp. 1935. 81 p.

[8] István Harsányi: A Sárospataki Rákóczi Könyvtár és Katalógusa (The Rákóczi Library at Sárospatak, and its catalogue). Bp. 1917. 3., 11., 22 p.

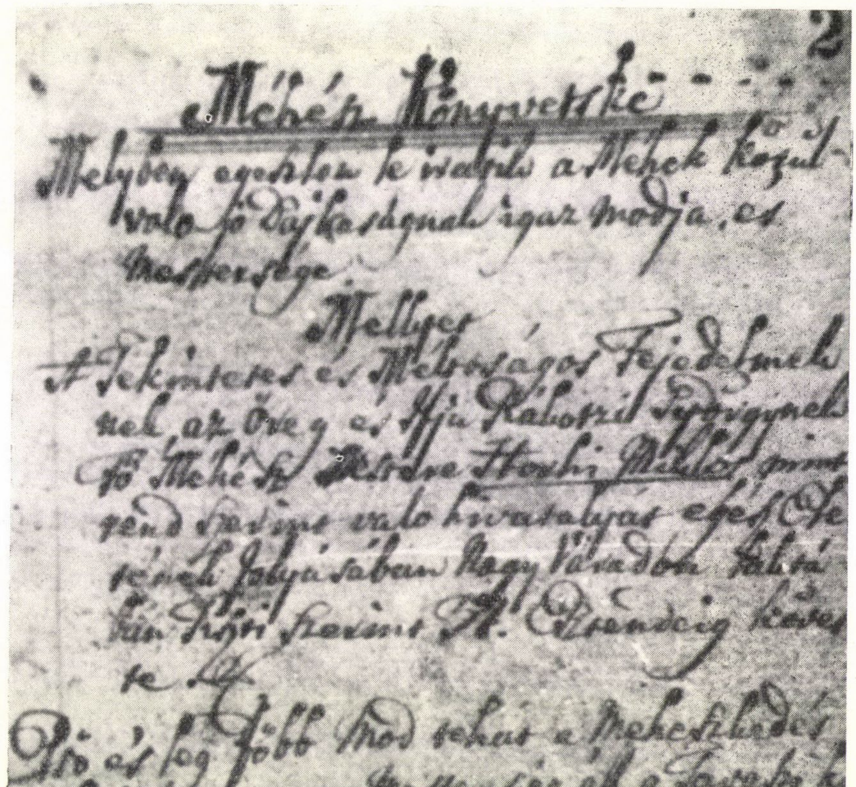

Fig. 1. First page of a handwritten copy made in 1720. Manuscripts of the National Széchényi Library. Call number: Quart. Hung. 2327

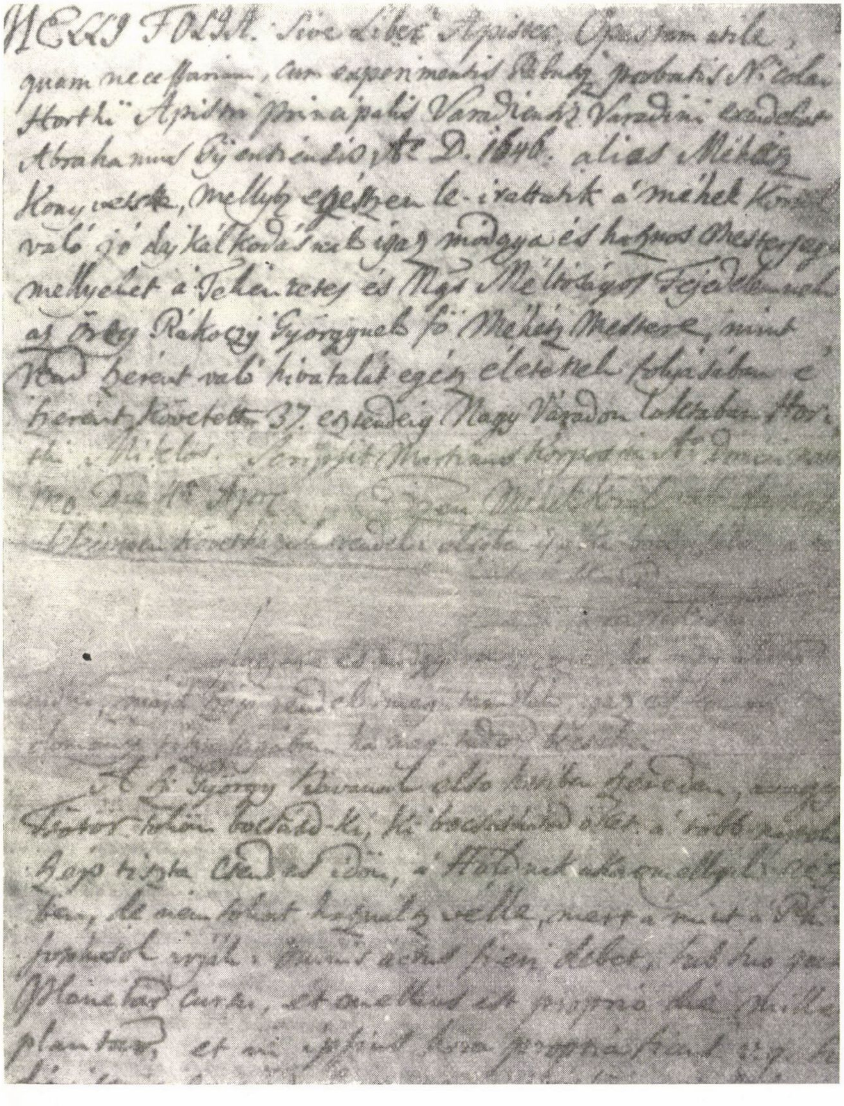

Fig. 2. First page of a handwritten copy made in 1802. Manuscripts of the National Széchényi Library. Call number: Oct. Hung. 1076

on March 25th, 1802 at Felsőörs. Ten years later, Jenő Rodiczky dr. published it in his book: On the bee . . . after an original hand-written copy in his possession. These two printed copies are totally identical except for some unimportant differences in letters. It was these two printed copies which were used by the author of the present paper.

Horhi's work was a practical handbook. All he wrote was based on the experiences of a whole life. He was no longer young when he wrote his book; he had already spent 37 years in taking care of the Prince's apiaries. At that time honey production was of considerable national economic importance which is proved by the fact that beside the trade of "szür"* and oxen, trading with honey and beeswax was also monopolized by György Rákóczi I. in 1631.[9]

Horhi's basic principle was the following saying of the ancients: "*Omnis actus et operatio debet fieri sub suo Planeta, et melius est propria Die in illius Planeta et ipsius hora*". That is: All actions and operations have to take place under their own planets, and are better performed on the proper day, under their own planets and in the right hour. Consequently, "the bees were put out ceremonially in spring" — as Zoltán Örösi Pál aptly remarks.[10]

There are superstitions in other parts of the "Apiarist booklet" as well, though not more than in other books published abroad even 100 years later. But interesting and clever apicultural techniques can also be found in it. Some of them are used even today.

In Horhi's days practical apiculture centred in the swarming of bees. It is thus understandable that the book deals with the control of swarming, collecting of swarms and co-ordination of swarming and honey production. The book recommends ringing or violin music as means of settling the swarms. It is very old though useless international practice. However the advice to sprinkle water with a leafy branch onto swarms leaving the hives is valid even today. The suggestion of scenting the hive with cumin flowers, cumin leaves or pine-resin prior to placing the swarm in is similarly useful. Excess swarming may be harmful, the colony becomes depopulated. To avoid it Horhi — quite rightly — recommends the removal of the queen-bee's cell: " . . . take the house of the queen-bee out". (The house of the queen-bee is the cell of the honeycomb where the queen-bee develops.) It is a widely used method against swarming even today.

As to the co-ordination of swarming and honey production Horhi recommends: "If you want to get much honey, after the bees have swarmed enough remove the honey often, as soon as the bees have filled the cells". Prevention of swarming by the removal of honey is well-known even today. Frequent honey extraction is recommended for the increase of honey production.

Robbery (bees attacking another hive to take honey away), was often dealt with by the literature of old times. Horhi threw poppy seed or sand among the fighting bees, so they were occupied with cleaning it away. It is a method recommended in essentials later too, and applied again nowadays.[11] Horhi suggested further to poison the attacking bees with honey mixed with yeast or stramonium. It was an international method frequently used both before and long after Horhi.

Horhi mentioned various pests and diseases. From his description we recognize the worm of the wax moth (he wrote about "wormy" bees). The meaning of the term "variolous bee" was found out from the explanations of later apicultural books. It is the phenomenon of worker's cells used as brood-cells and covered with small-pox-like blisters (in case the queen-bee is unfit or absent, and oviposition is carried out by the workers). Horhi recommends salted wine or brandy against the diseases. At that time and long after nothing wiser was known even abroad.

He recommends — like the ancient Roman and mediaeval authors — boiled sweet plums, dried grapes, raisins, roast chicken, just the same as abroad at that time.[12]

Finally, two further interesting points should be mentioned here. Horhi left those bee colonies overwinter which around St. George's Day flew "more vigourously", that is, more frequently than the others, so were thought to stand winter better. And he did not kill the bees (e.g. with sulphur, fume or fuzz-ball) to get the honey, only removed the filled honeycomb.

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[8] Long embroidered felt cloak of Hungarian shepherd.

[9] Rodiczky dr.: A Méhről . . . (On the bee . . .) pp. 72—73.

[10] Méhek között (Among bees). Bp. 1951. 519 p.

[11] See the review published on page 56. of No. 3. 1970. of the journal "Méhészet".

[12] Zoltán Örösi Pál op. cit. 519. p.

He definitely wrote: "Don't kill the bees . . .". He laid down a principle of great importance, since in Hungary the destruction of healthy bee-colonies was only forbidden by the decree No. 66.113/1947. F.M.

It should not be forgotten that it was Nicolaus Horhi who with his "Apiarist booklet" started the literature of apiculture in Hungary and was for more than a hundred years, up to the middle of the 18th century its only representative. The next work on apiculture, "Ángliai méhes kert" (Apiaries in England) by János Gedde was translated by György Szattmáry Király and published in Hungarian in 1759 in Eger. Accordingly, Horhi's *Liber Apiaster* is valued with Vergilius' words: "The work — though mediocre — is not without honour".[13]

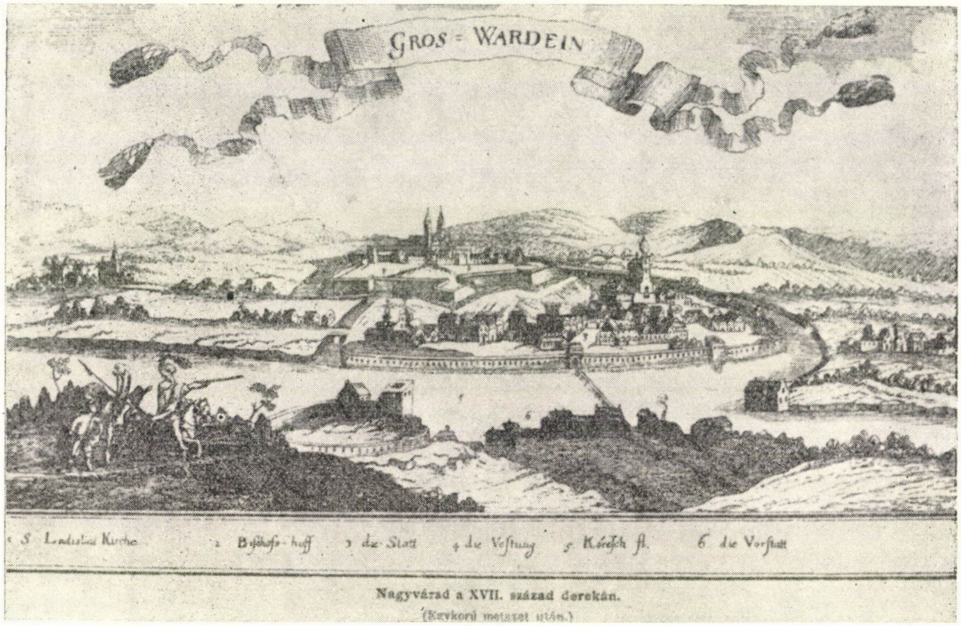

Fig. 3. Nagyvárad in the middle of the 17th century. After a contemporary print. Ignácz Acsády: Magyarország Története (History of Hungary). Bp. 1898. p. 81

The cause of disappearance of Horhi's book. Let us give here a short explanation of how it could happen that "this interesting literary relic" has totally disappeared and not a single original printed copy is known to us. The cause of its disappearance is to be searched in the stormy history of Hungary and the Hungarian people.

It was already during the principality of György Rákóczi II. (1621—1660) that the glory of Transylvania declined. Namely, in 1657 the Prince — without the approval of the Turks, moreover, in spite of the protestation of the Porte — led a campaign to occupy the Polish throne. He was, however, defeated and his army captured by the Tartars. The Ottoman Porte turned against the ambitious and disobedient vassal, and the "rebellous" Transylvania was raided by Turkish and Tartar forces.

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[13] Georgicon IV. 6.

Nagyvárad (town in Transylvania) where in the printing shop of Ábrahám Szenczi Kertész, the excellent printer of the 17th century, among 70 Hungarian publications the "Apiarist booklet" was published, also fell into the hands of the Turks in 1660.[14]

The disappearance of the book is explained further by the following: Zsófia Báthory (1629–1680) wife of György Rákóczi II. after her husband's death returned to the Catholic Church, invited Jesuits to the Court and on October 20th, 1671 expelled the teachers and students of the College from Sárospatak. The books of the Library became the possession of the Jesuits, except those taken by the teachers, János Pósaházi and Mihály Buzinkai, and the students to Debrecen, then to Gyulafehérvár and Marosvásárhely. Meanwhile even the books left behind at Sárospatak changed hand several times, with the town itself.[15]

Fig. 4. György Rákóczi I. Portrait from the 17th century in the Hungarian Historical Picture Gallery. Dávid Angyal: Magyarország Története (History of Hungary). Bp. 1898. p. 488

The situation was similar in the other parts of the country too. In 1686 Buda was released from Turkish occupation. Hungary, as a battlefield, made great sacrifices during the war. After the Turks the country was raided by the army of the German Kaiser. Finally, exasperation and discontentedness broke out in Ferenc Rákóczi II.'s (1676–1735) war of independence. So the following decades were again bloody and ruthless on account of a political

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[14] Andor Tevan: A könyv évezredes útja (Thousand years history of the book). Bp. 1956. 202 p. — József Fitz: A magyar könyv története 1711-ig (History of the Hungarian book up to 1711). Bp. 1959. pp. 155–156.

[15] István Harsányi op. cit. pp. 23–25.

despair and religious fanaticism. After the defeat of the war of independence neither people nor books were spared. No wonder if books were found "in the walls of desolate churches". Of the 821 volumes enlisted in the catalogue of the famous Rákóczi Library at Sárospatak there are ten books of which not a single copy has been found, and even their existence is proved only by this catalogue.[16]

And as the following century is best characterized by the poet's words: "There has been a great storm. No peaceful hiding place will you find in our desolate groves",[17] — my study will be completed with a wish of nearly a hundred years old: "May a printed copy of Nicolaus Horhi's Liber Apiaster be discovered".[18] We greatly regret the disappearance of the book, and are indebted to those nameless persons who by their handwritten copies have preserved the teaching of the first Hungarian book on apiculture for us.

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Zoltán Örösi Pál, leader of the Research Institute for Breeding Small Animals, Gödöllő, gave substantial assistance for the presentation of Horhi's "Apiarist booklet". Here again, I thank him for his expert advice and guidance.

P. Hargita

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INVESTIGATIONS INTO COMBINE HARVESTER CROP CHARACTERISTICS IN TERMS OF AGROBIOLOGY, PLANT PHYSIOLOGY AND MORPHOLOGY, AND APPLICATION OF RESULTS TO ACHIEVE OPTIMUM TECHNOLOGICAL AND TECHNICAL CONDITIONS IN COMBINE HARVESTER OPERATION

II. OPTIMUM COMBINE HARVESTER SETTING AND IMMEDIATE LOSS DETERMINATION

1. Application of data and experience acquired in plant investigations for optimization of combine harvester operation

When the data and insight acquired in the course of the investigations described in Part I were assessed and evaluated the obvious point was to apply to practical farming the setting data for the combine harvester operating elements that were established in the tech-

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[16] Ibid. on pages 22. and 23. see the titles and authors of the ten lost books enlisted in detail.

[17] Mihály Tompa (1817—1868): A madár fiaihoz (The bird's song to her nestlings). 1852.

[18] Károly Szabó: Régi magyar könyvészeti adalékok (Data on old Hungarian books). Magyar Könyv-Szemle. Bp. 1879. 121 p. Horhi's book is mentioned by Grossinger, historia Physica Regni Hungariae. Tom. IV. 51. — Hiador Sztripszky dr.: Adalékok Szabó Károly Régi Magyar Könyvtár I.—II. kötetéhez (Contribution to Károly Szabó's Old Hungarian Library I.—II.). Bp. 1912. 144 p.

nical test. The setting data required for optimum efficiency and threshed grain quality dependent on moisture content and ripening conditions have therefore been established for every combine harvester crop involved.

A combine harvester setting disk and a combine harvester setting slide rule have been designed in combination described elsewhere.

1.1. Setting slide rule (machine setting and loss determination).

The setting slide rule provides combine harvester operators with the data required for optimum machine setting, covering the commonly grown crops in a systematic and easily readable way. The setting data are indicated for each crop involved, arranged according to harvesting conditions and dependent on moisture content (Fig. 1).

The data required for immediate loss determination have also been entered there to allow quick checking of the machine setting applied (Fig. 2). In order to meet all the harvesting conditions involved as far as possible in the case of each crop the setting data for three different harvesting conditions are indicated.

(a) Dry = straw brittle/grain moisture content 12 to 14%
(b) Medium = straw of medium strength /grain moisture content 15 to 17%
(c) Moist = straw tough/grain moisture content 18 to 20%

Slide rule rear serves for immediate loss determination (Fig. 8).

1.2. Setting slide rule application.

In order to find out the setting data the slide rule user need only set the reading window on the crop involved and can then read the setting parameters dependent on grain moisture content.

In field work the following procedure is recommended for optimum combine harvester adjustment with the aid of the setting slide rule:

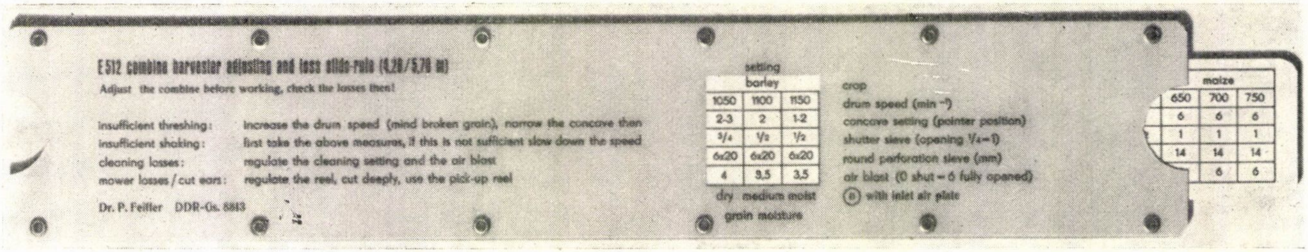
(a) Assessment of grain suitability for threshing: fall-out test
grain seating check
moisture content check
(b) Selection of combine harvester setting data according to harvesting conditions involved
(c) Setting of combine harvester operating elements
(d) Threshing trial run
(e) Immediate loss determination to check machine setting
(f) Readjustment of machine setting in case of inadmissible threshing losses.

1.3. Optimization of setting.

1.3.1. Assessment of grain suitability for threshing. The assessment of the ripening level and an accurate moisture content measurement are the necessary conditions for a proper combine harvester setting. These characteristics indicate grain suitability for threshing.

Apart from the assessed threshing suitability the other indices required for the definition of the most preferable minimum-loss harvesting season are related to terminal ripeness. The following three checks have proved useful:

(a) Manual handling
(b) Assessment of fall-out losses (Fig. 3)



E 512 combine harvester adjusting and loss slide-rule (4,20/5,70 m)

Adjust the combine before working, check the losses then!

insufficient threshing: increase the drum speed (mind broken grain), narrow the concave then

insufficient shaking: first take the above measures, if this is not sufficient slow down the speed

cleaning losses: regulate the cleaning setting and the air blast

mower losses/cut ears: regulate the reel, cut deeply, use the pick-up reel

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setting

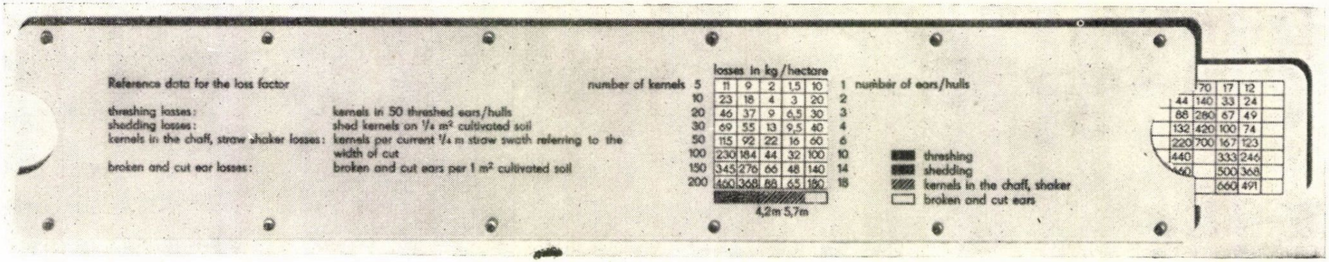
barley			maize			crop
1050	1100	1150	650	700	750	drum speed (min⁻¹)
2-3	2	1-2	6	6	6	concave setting (pointer position)
³/₄	¹/₂	¹/₂	1	1	1	shutter sieve (opening ¹/₄–1)
6x20	6x20	6x20	14	14	14	round perforation sieve (mm)
4	3,5	3,5		6	6	air blast (0 shut – 6 fully opened)

dry medium moist

grain moisture

Ⓢ with inlet air plate

Fig. 1. Setting slide rule front



Reference data for the loss factor

threshing losses: kernels in 50 threshed ears/hulls

shedding losses: shed kernels on ¹/₄ m² cultivated soil

kernels in the chaff, straw shaker losses: kernels per current ¹/₄ m straw swath referring to the width of cut

broken and cut ear losses: broken and cut ears per 1 m² cultivated soil

losses in kg/hectare

number of kernels						number of ears/hulls
5	11	9	2	1,5	10	1
10	23	18	4	3	20	2
20	46	37	9	6,5	30	3
30	69	55	13	9,5	40	4
50	115	92	22	16	60	6
100	230	184	44	32	100	10
150	345	276	66	48	140	14
200	460	368	88	65	180	18

4,2m 5,7m

- ■ threshing
- ■ shedding
- ▨ kernels in the chaff, shaker
- ☐ broken and cut ears

	70	17	12	
44	140	33	24	
88	280	67	49	
132	420	100	74	
220	700	167	123	
440		333	246	
660		500	368	
		660	491	

Fig. 2. Setting slide rule rear

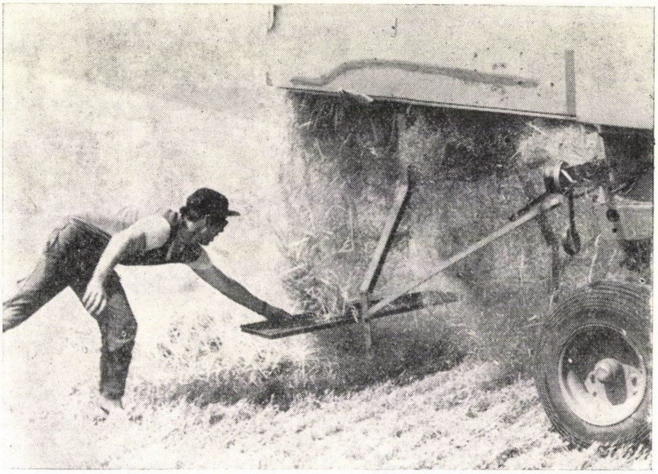


Fig. 3. Check bowl being placed below the falling straw swath to determine shaker and scattering losses

Fig. 4. Straw resting above the check bowl is shaken to make residual grain drop into the bowl

(c) Assessment of drum losses (Figs 4, 5)

(d) General rules applied to assess threshing suitability

- Gran should be hard so that it cannot be broken by the thumb;
- Grain cracks audibly when torn (moisture content below 15 per cent), oats may be somewhat softer;
- Grain has lost its gloss, barley in particular looks dull and wrinkled.

Fig. 5. A lot of grains are in the bowl. In this case the slide rule in the column dealing with shaker and scattering losses will indicate a loss rate of more than 65 kg/ha

1.4. Definition and classification of threshing suitability.

Three moisture-specific subdivisions are indicated on the setting slide rule to define and classify grain threshing suitability.

(a) "dry"

= grain well suited for threshing
= loose to medium-tight grain seating
= grain moisture content of 12 to 14 per cent

The term "dry" indicates a powdery straw that can be chopped fairly easily (Fig. 6), easily bursting husks or hulls, a faster filling of the screens and shakers by the resulting higher proportion of chaff.

The grain has attained a highly suitable or at least suitable level of terminal ripeness.

The ears are in a slightly crisp condition, the grains will relatively easily be released during threshing. Even when affected to a greater extent by rain or dew the straw loses much of its flexibility so that drum winding is not to be expected any more.

Crops that have reached full terminal ripeness are very well or fairly well suited for threshing. Fall-out is about to begin, and both straw and stem elements are free of water.

(b) "moist"

= grain rather unsuitable for threshing
= medium-tight to tight grain seating
= grain moisture content of 18 to 20 per cent

The term "moist" indicates a condition in which the crop is moist to some degree both outside and inside, with easily winding straw and with hulls and husks that will resist bursting during threshing, with water-repellent tough stems; the grains and seeds will resist being released from the ears and panicles. Independent of the momentary weather conditions involved the level of crop ripeness will just barely allow efficient threshing. There is also still water in the lower conduits of the straw even if there is a dry spell, i.e. independent of momentary weather conditions. The straw is decidedly tough and resists being torn. Heavy beating is required to release all the grains from the ears.

In general therefore all varieties show slight to medium suitability for threshing shortly after or when attaining threshing ripeness. This slight to medium suitability applies in the entire harvesting season to some of the beard-fast varieties or those with extremely tight grain seating, too.

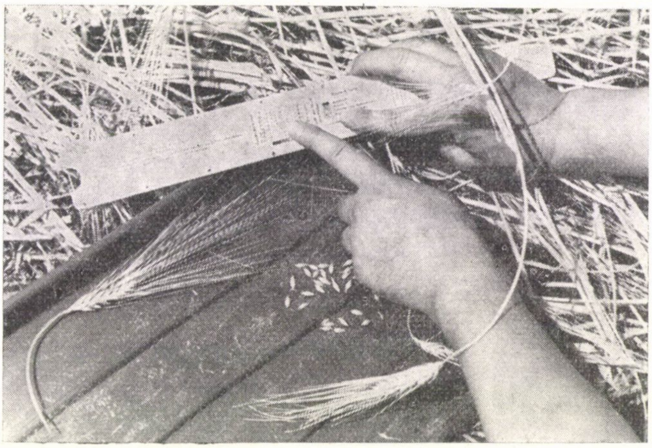


Fig. 6. After adjustment of combine harvester setting and another check bowl test there are only 5 grains in the bowl. The slide rule in its column dealing with shaker and scattering losses shows that the loss rate is low, amounting to 1.5 kg/ha only

Fig. 7. Checking of fall-out loss before threshing. Fall-out rate is particularly high with overripe crops

(c) "medium"

= medium suitability for threshing
= medium-tight grain seating
= grain moisture content of 15 to 17 per cent

This term indicates average harvesting conditions. Setting data from this range should always be chosen when decisive factors (i.e. moisture content, ripeness level, etc.) do not correspond to the other two ranges mentioned above.

Terms "dry", "medium", "moist" are also the only ones used for non-grain crops. Their harvest season is short and their complete drying, particularly the more and more widely applied defoliation, does not require any more distinction. In addition, the threshing date will have to be carefully selected taking crop brittleness and the date at which the chemical defoliating agent had been sprayed into consideration.

Crop conditions can preferably be evaluated by the following general rule:

Grain has attained high suitability for threshing when fall-out involves more than 20 grains per square metre, when the straw moisture content is below 30 per cent, or when more than 3 buckling ears per square metre can be found.

Grain has a low level of threshing suitability when fall-out involves less than 10 grains per square metre, when there is less than one buckling ear per square metre, and when the straw moisture content exceeds 30 per cent. Attention should be paid to the latter index since it will lead to an increase of grain moisture content during the threshing operation, with moisture increasing by 1 to 3 per cent. It should be kept in mind when moisture content conditions are considered for storage and drying.

1.5. Combine harvester setting principles

The following principles are to be applied in combine harvester setting or setting parameter selection:

Grain of high moisture content and low threshing suitability, i.e. shortly after attaining terminal ripeness, after heavy rain or dew, "keen" threshing is necessary. In this moisture-content or ripening condition, respectively, the grain is less liable to break, and arrangements can therefore provide for "keener" threshing.

The condition of the straw, which has just ripened and often is still slightly green in its lower sections, complicates the work of the threshing gear and shaker and also requires rigorous action by the threshing elements.

A higher threshing drum speed and a slightly narrower cage are therefore required for maximum threshing yield and efficient separation. The combine harvester will thus have to move more slowly, or the feed will have to be reduced.

This in turn calls for a slightly relaxed setting of the cleaning elements.

The following conclusions were reached:

Moist grain = slightly slower combine harvester travel (compared to identical dry crop density)
= slightly higher threshing drum speed
= sligthly narrower cage setting
= slightly less wind and/or closer screens

It is just the reverse with dry grain. Breaking hazards increase. The drum speed must not be as high as stated above. A high drum speed is of course not even required as at that moisture-content or ripeness level the grain will easily be released from the ears and panicles.

The throughput or feed, i.e. combine harvester travel speed, can thus be increased.

To ensure the proper processing of the increased feed the cage will have to be set more widely. The more compact flow of grain will provide an additional protection for the brittle

constituents. Higher throughput rates and higher chaff shares of course require slightly more wind and wider screens, respectively. There is an optimum interrelationship between all the setting indices involved.

Fig. 8. Handling of threshed ears to determine drum losses

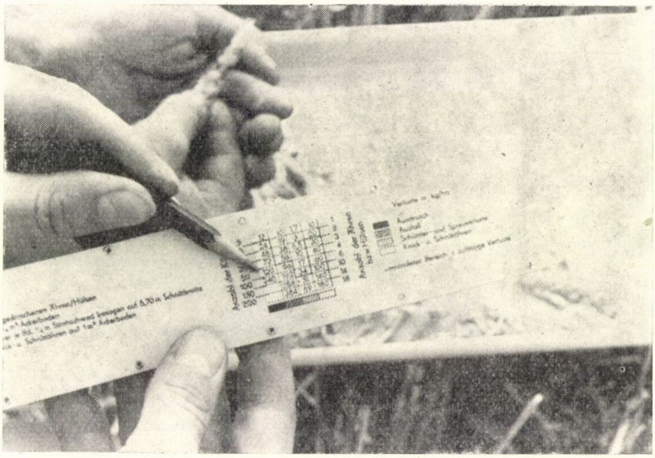


Fig. 9. There are many grains (i.e. more than 50) in the 50 threshed ears involved. This indicates that drum loss is high

The following conclusions were reached:

Dry grain = slightly faster combine harvester travel (compared to identical moist crop density)
= slightly lower threshing drum speed
= slightly wider cage setting
= slitghly more wind and/or wider screens

This survey roughly represents the system of moisture-content and ripening level consideration applied in the selection of the setting data and the throughput-specific optimum combine harvester setting.

The system combines optimum efficiency, minimum losses, and the most favourable quality indices.

1.6. Loss and quality level checks and adequate setting adjustment

Establish grain loss (in reading window, from left to right).

1st step: Check threshing or drum losses, i.e. amount of grains remaining in the ears.

Pick at random 50 threshed ears from the straw swath, remove any grains still contained, count them.

Repeat the above procedure three times. Enter the number of grains thus established (e.g. in case of wheat 20 grains per 50 threshed ears = 48 kg/ha) as loss in the first column (from left to right) in the reading window.

2nd step: Count the number of fall-out and scattering loss grains per 0.25 m^2 of crop area before and after threshing.

(Example: Prior to threshing with 3 counts an average number of 20 grains = 38 kg/ha loss due to fall-out; after threshing or mowing, resp., an average number of 40 grains = additional 38 kg/ha loss due to actions of reel and other cutting elements).

Enter the number of grains thus established in the second column in the reading window.

3rd step: Check combined shaker and chaff losses, which may well be the most important source of losses, or separately check the losses from the shaker and chaff.

Fig. 10. In the field there are about 20 to 50 stems twisted to find out which setting to select, i.e. dry, medium, or moist. Since straw in this case is crisp and brittle, "dry" is the condition selected

Place a check bowl or something answering the purpose of 25-cm width below the falling straw swath and/or chaff outlet. Remove straw, blow off the chaff, count the number of grains caught.

Repeat the above procedure three to five times. Enter the average number of lost grains (e.g. 200 grains caught in bowl = 69 kg/ha loss) in the third column in the reading window. (If there is not any suitable vessel available it may be sufficient to count the number of grains exhibiting fresh gloss below a straw swath area of 25 cm.)

4th step: Ears broken or cut off (These data involve the fourth (last) loss column; the first three apply to grains.) Count the number of ears found on 1 m^2 of crop area before (i.e. ears damaged by breaking or buckling = buckling ears) and after (cut-off ears) threshing. Execute this procedure three to five times before and after threshing.

(Example: 5 counts before threshing in case of winter wheat resulting in an average of 3 ears/m^2 = 30 kg/ha loss by ears damaged by breaking or buckling; while counts after threshing or moving, resp., result in an average of 5 ears/m^2 = an additional loss of 20 kg/ha due to cut-off ears).

5th step: Proceed with established losses of grains and ears (panicles, hulls/husks from the left-hand or, resp., the right-hand side of the reading window on the rear of the setting slide rule, taking into consideration the 4 differently marked rectangles at the lower edge of the reading window, and establish the level of the 4 different grain losses in terms of kg/ha. Taking our examples given in steps 1 to 4 we arrive at the following result:

(a) 20 grains per 50 ears not released in threshing = 48 kg/ha loss
(b) 20 grains per 0.25 m^2 due to fall-out = 38 kg/ha loss
and
20 grains per 0.25 m^2 due to scattering = 38 kg/ha loss
(c) 200 grains per 25 cm of straw swath = 69 kg/ha loss
(d) 3 ears per 1 m^2 crop area damaged by buckling = 30 kg/ha loss
and
2 ears per 1 m^2 crop area damaged by cutting = 20 kg/ha loss
Total loss = 243 kg/ha

6th step: Check whether the loss rate resulting from the four different sources is within admissible limits; add total losses and state result in terms of kg/ha and percentage of the estimated rough yield.

(When we proceed from an estimated winter wheat yield of 50 dt/ha the loss of 243 kg/ha given in our example corresponds to about 5 per cent loss of actual yield).

Steps 1 to 6 clearly demonstrate to which level the loss rate may rise even with modern combine harvesters if losses are not checked thoroughly.

International reference literature on combine harvesters giving loss rates of 5 to 10 per cent (or even higher in case of non-grain crops) proves that loss rates are actually in this range.

7th step: Draw conclusions from steps 5 and 6 and decide whether combine harvester setting should be adjusted or not; adjustment will usually be required.

Reduce grain losses, according to the information as to causes provided on the front of the setting slide rule, one after the other by adjusting the various combine harvester operating elements involved.

In our example increase drum speed and narrow the cage arrangement, this will markedly reduce threshing yield losses.

Whenever any adjustment is made, i.e. here the increase of drum speed and the narrowing of the cage gap, with a resulting exacerbation of threshing intensity it is very important to check the proportion of broken grain.

Threshing intensity may be exacerbated only as far as grain damage will allow.

Shaker losses will slightly decrease due to improved threshing yield and more favourable (i.e. earlier) separation.

Adjust reel speed — with resulting decrease of scattering losses.

Drive more slowly if need be, this will result in a more marked decrease of shaker and scattering losses, the cutting level can be set lower and losses due to cut-off ears will also decrease.

This allows losses due to threshing conditions (i.e. grain not released in threshing, scattering loss, discharge of loose grain together with straw and chaff, cut-off ears) to be reduced to the required level of 1.5 per cent from yield.

Fall-out losses and losses from buckling ears, which are due to plant and ripening conditions, will have to be continuously checked for days before harvesting starts, and a harvesting operation sequence will have to be laid down in view of the established data in order to keep these losses as low as possible.

8th step: Determine final grain losses and enter the resulting data in the harvesting diary separately according to types of grain loss and as total loss in terms of kg/ha and percentage of overall yield. If required execute final adjustment of machine setting.

In compliance with changing threshing conditions (e.g. drier weather, different ripening level, lower moisture-content level, etc.) check losses several times a day and accordingly adjust machine setting and vary combine harvester travel speed (which may increase still more in the course of the day if drying continues).

2. Economic benefit

Investigations carried out in recent years show that application of optimum setting data resulted in considerable economic benefits. In the period ranging from 1963 to 1966 harvesting losses could be reduced down to 2 per cent with grain crops and to 8 per cent with non-grain crops, related to the total yield. Efficiency increases attained at the same time ranged from 17 to 22 per cent.

Compared to harvesting losses of 10 per cent and more stated in reference literature this is a most remarkable result.

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STUDY ON THE PHYSIOLOGICAL SPECIALIZATION OF ERYSIPHE GRAMINIS DC F.SP. TRICITI MARCHAL AT MARTONVÁSÁR 1970/71

In the winter of 1970 an opportunity arose to extend research work concerning the powdery mildew of wheat and deal more intensively with the pathogen. A part of this program was to assess the race spectre of powdery mildew and study the attitudes of the varieties towards the various races.

The first step of our work was of an assessing character. For this reason pure cultures of numerous hybrids and varieties were isolated.

Our investigations were carried out in a green-house during the winter period at Martonvásár, at the Agricultural Research Institute of the Hungarian Academy of Sciences. During the experiment the temperature was 16—20°C, relative humidity was 60—80 per cent. Artificial illumination was also applied when required.

Race determination was carried out with NOVER's (1957) method modified to some extent by ourselves (SZUNICS 1969). Reaction of the varieties to various races was evaluated by our method described earlier (SZUNICS 1969), while resistance to the population (susceptibility) according to methodics recommended by HINFNER (1960), with artificial infection applied in each case.

Table 1

Percentage distribution of race spectre

(Martonvásár, 1970/71)

Isolated races	Frequency		Isolated races	Frequency	
	number	percentage		number	percentage
0	5	10.0	16	2	4.0
2	1	2.0	18	1	2.0
3	7	14.0	24	1	2.0
4	1	2.0	26	1	2.0
5	4	8.0	27	2	4.0
7	3	6.0	31	1	2.0
9	7	14.0	32	2	4.0
13	2	4.0	35	4	8.0
14	1	2.0	X	5	10.0

$$\Sigma \text{ isolate} = 50$$

$$\Sigma \text{ race} = 18$$

$$\frac{\Sigma \text{ isolate}}{\Sigma \text{ race}} = 2.77$$

$$\frac{\Sigma \text{ race}}{\Sigma \text{ isolate}} = 0.36$$

Powdery mildew was collected from different varieties and hybrid material sown in the experimental area.

a) Assessment of the race spectre. On the basis of fifty pure lines 18 races were identified (Table 1). Races most frequently occurring in the relatively small population are: 3; 9; 0. Races 3 and 0 are present in many countries of Europe. In England they are the dominant races (WOLFE 1967). Race 9 was isolated — besides Hungary — in the German Democratic Republic (NOVER 1957), Yugoslavia (SMILJAKOVIC 1966) and Italy (GRASSO 1966).

Of races 16, 18 and 19 domineering in Krasnodar race 19 could not be isolated in the first year. This is all the more interesting because all the three races occur in the variety Berostaya 1 (SZUNICS 1969).

In the population examined a new race not yet described was isolated (marked provisionally with X), which, at the same time, proved to be the most agressive and infected the following varieties tested: Carsten V.; Salzmünde Stamm 1444; Red Fern; Axminster; Normandie; Hope; Chul. (Table 2).

Resistance to races 9, 35 and X was examined in eight varieties in Hungary. Table 2 shows the agressivity of these races. The pathological picture of a race isolated in Krasnodar in 1969 and labelled with 45 by Nover (personal publication in 1971) is found in the same Table.

b) Resistance of some varieties to the various races of powdery mildew. From the point of view of breeding work and production it is important to know not only the races which the local population consists of but also which of them are the most agressive as well as the dominant ones, and how the commercially produced varieties react to these races.

Table 2

Reaction of varieties tested to four different races

Varieties tested	Races			
	9	35	45	X
Carsten V	S	S	S	S
Salzmünde Stamm 14/44	S	S	S	S
Red Fern	S	R	R	S
Axminster	S	S	S	S
Normandie	S	R	R	S
Halle Stamm 13471	R	R	R	R
Weihenstephan M I	R	R	S	R
Hope	R	S	R	S
Chul	R	S	R	S

Note: R = resistant
S = susceptible

Table 3 shows the powdery mildew resistance of several varieties to the Martonvásár population and to races 9, 35 and X.

The varieties Avrora and Kavkaz are considered populations from the point of view of powdery mildew resistance. Consequently, it may be useful to bring them to an immunological balance, and use the resistant form obtained in this way in commercial production and as a basic material of breeding. It may be all the more useful, as these varieties are heterozygous in relation to race X too (they segregate), moreover, as a reaction to the pathogen, intensive necrotic and chlorotic patches can be found on the plants. The other varieties examined are totally susceptible to the mentioned race.

The extent of resistance to various races and to the population do not contradict each other. Nevertheless, evaluation of resistance to the various races gives a much more detailed result, since some varieties do not have the same susceptibility, or resistance to the different races.

Table 4 contains the material of which the pure lines were isolated.

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Prepared at the Agricultural Research Institute of the Hungarian Academy of Sciences, Martonvásár

L. Szunics, L. Szunics

Table 3

Resistance of several varieties grown in Hungary to races 9, 35 and "X", and to the local population

(Martonvásár, 1970/71)

Variety	Races						Population
	9		35		X		
	type of infection	infection percentage	type of infection	infection percentage	type of infection	infection percentage	infection percentage
Avrora	0—3 cN	5.3 (0.0—50.0)	0—3	2.9 (0.0—10.0)	0—4 CN	11.6 (0.1—30.0)	6.2 (0.0—60.0)
Kavkaz	0—1 C	0.01 (0.0— 0.1)	0—2	0.95 (0.0— 5.0)	0—3 C	9.02 (0.1—50.0)	7.8 (0.0—60.0)
Fertődi 293	0—4	51.4 (0.1—60.0)	1—4	60.5	4	90.0	83.2
Mir. Jub. 50	2—4	60.5 (2.0—70.0)	4	95.0	4	95.0	85.0
Bezostaya 1	4	60.0	4	90.0	4	90.0	87.5
Mironovskaya 808	2—4	28.2 (3.0—40.0)	4	90.0	4	100.0	87.5
Rannaya 12	3—4	40.0	4 c	70.0	4	90.0	92.5
Moisson	2—3	70.0	4	85.0	4	100.0	100.0

Note: C = intensive chlorosis
c = slight chlorosis
N = large necrotic patches

In some places two figures are given in the columns of infection type and infection percentage; this means that the variety does not uniformly react to the races indicated.

Table 4

Distribution of races isolated from various varieties and hybrid combinations in 1970/71

Varieties and hybrids	Isolated races	Number of isolated races
Hybrids	3, 4, 5, 9, 16, 18, X	7
Besostaya 1	0, 9, 26, 27, 32, 35	6
Fertődi 293	3, 7, 9, 14, 16	5
Avrora	3, 5, 24	3
Skorospelka 35	7, 9, 32	3
Mironovskaya 808	3, 9, 35	3
Kiszombori 1	5, 13, 35	3
Mv 69-13	0, 2, 13	3
Besostaya 1 dwarf	7, 9	2
Kavkaz	35, X	2
Skorospelka 3b	5, 31	2

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CHANGES IN THE ULTRASTRUCTURE OF POLARIZING MERISTEM CELLS UNDER THE INFLUENCE OF COLCHICIN

Differentiation in the meristem cells and tissues is caused fundamentally by polarity occurring in a certain phase of cell development. The consequence of polarity is an unequal cell division which results in different cell types and tissues (Bünning 1958, Bloch 1965). Colchicin inhibits the development of polarity in the meristem cells thus influencing the differentiation of cells and tissues to a great extent (Weissenböck 1949, 1950, Bünning—Hunck—Lutz 1965, Wettstein 1965).

Our investigations were aimed at getting information on the ultrastructural changes occurring in the polarizing cells of root tip meristems under the influence of colchicin. Young root tips of *Allium cepa* L. and *Zea mays* L. represented the object of investgation. In the experiments the seedlings were kept in 0.2—0.4 % colchicin solutions for 2—4 days; during this time a thickening characteristic of the influence of colchicin became apparent on the root tips. The cell structures of treated roots were compared with those of untreated, normal roots. For the purpose of electron microscope studies root tips were fixed in a 2 per cent buffered potassium permanganate solution, then — after having been washed out and dried — inbedded in araldite in the usual way. The ultra-thin sections were contrasted in plumbum citrate solution.

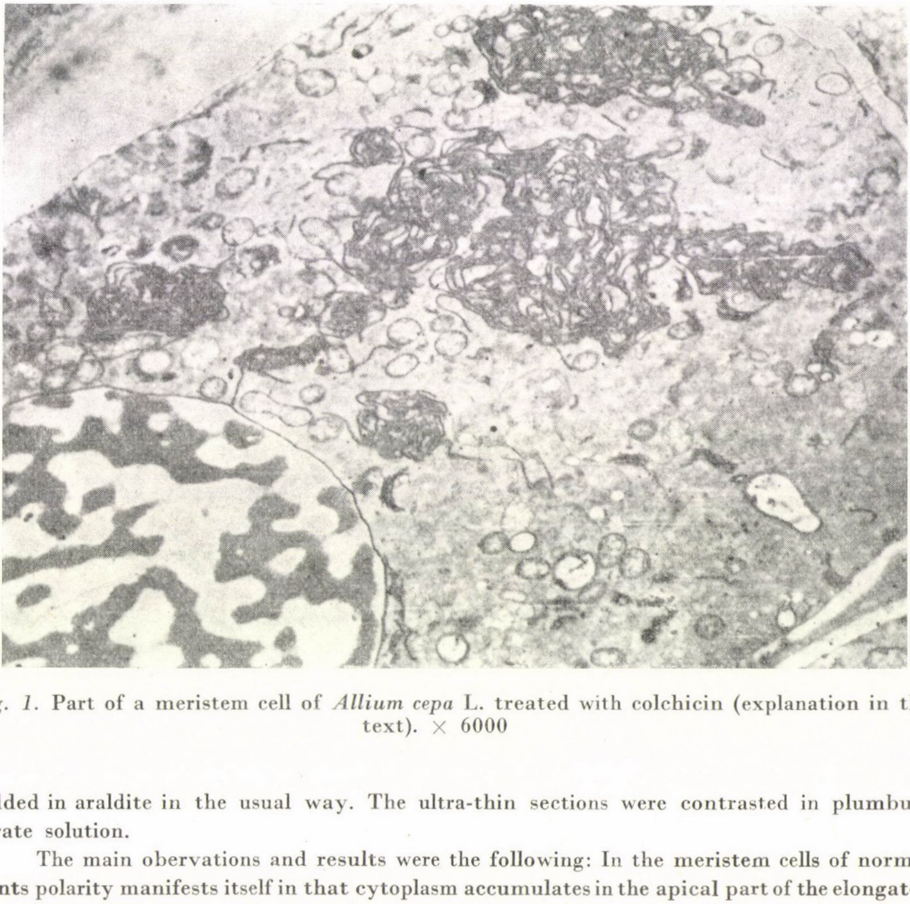

Fig. 1. Part of a meristem cell of *Allium cepa* L. treated with colchicin (explanation in the text). × 6000

The main obervations and results were the following: In the meristem cells of normal plants polarity manifests itself in that cytoplasm accumulates in the apical part of the elongated cell and cytoplasm organelles — primarily the endoplasmic reticulum (ER) — occurring there in larger quantities. Polarization does not occur in meristem cells which at the time of the colchicin treatment have not yet polarized, such cells grow isodiametrically and the ER disintegrates into smaller or larger vesicula. In cells affected by the colchicin treatment ultrastructural changes are of much higher extent at the stage of polarization. The most conspicuous phenomenon is the intensive accumulation of ER, which starts partly in the apical cytoplasm, partly in centres near the nucleus (Fig. 1), then — after constantly growing — becomes an unbroken system (Fig. 2). In cells already polarized a thick mass of ER can be found beside the nucleus, in the direction of the apical part of the cell (Fig. 3). Further, an increase in the number of mitochondria

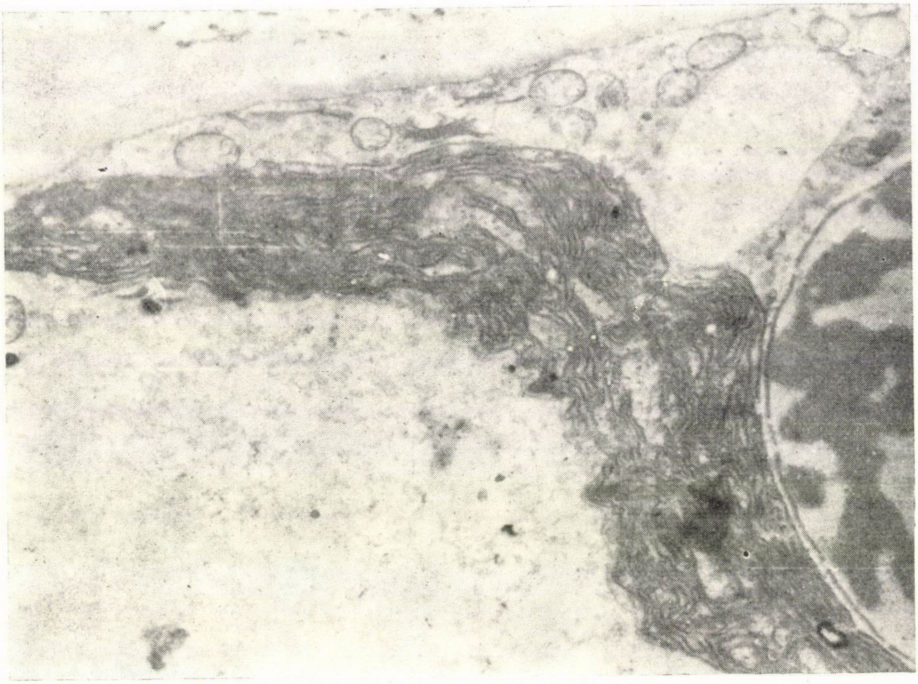

Fig. 2. Part of a meristem cell of *Allium cepa* L. treated with colchicin (explanation in the text). × 9000

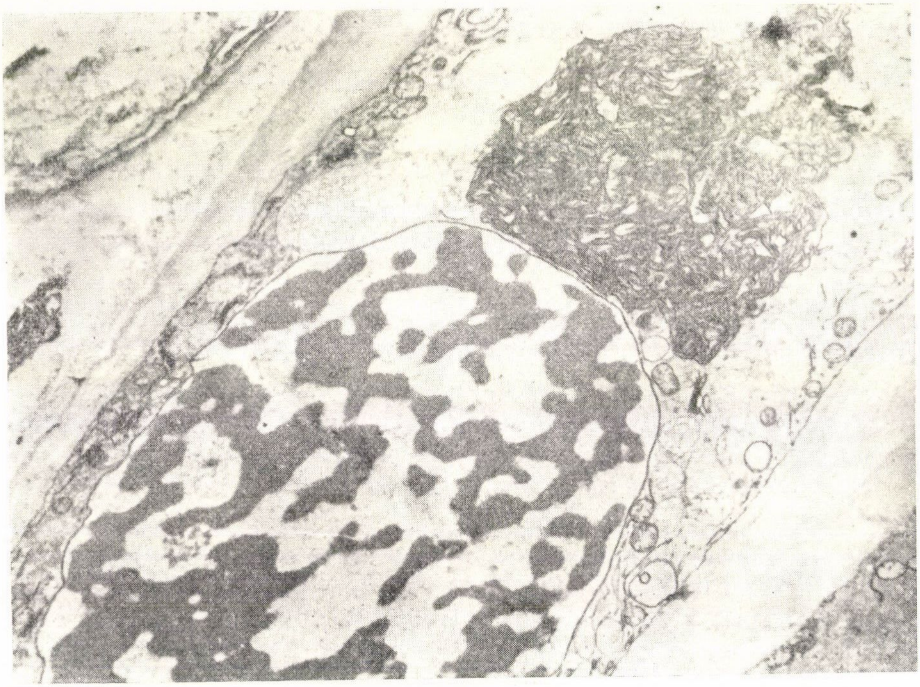

Fig. 3. Part of a meristem cell of *Allium cepa* L. treated with colchicin (explanation in the text). × 4500

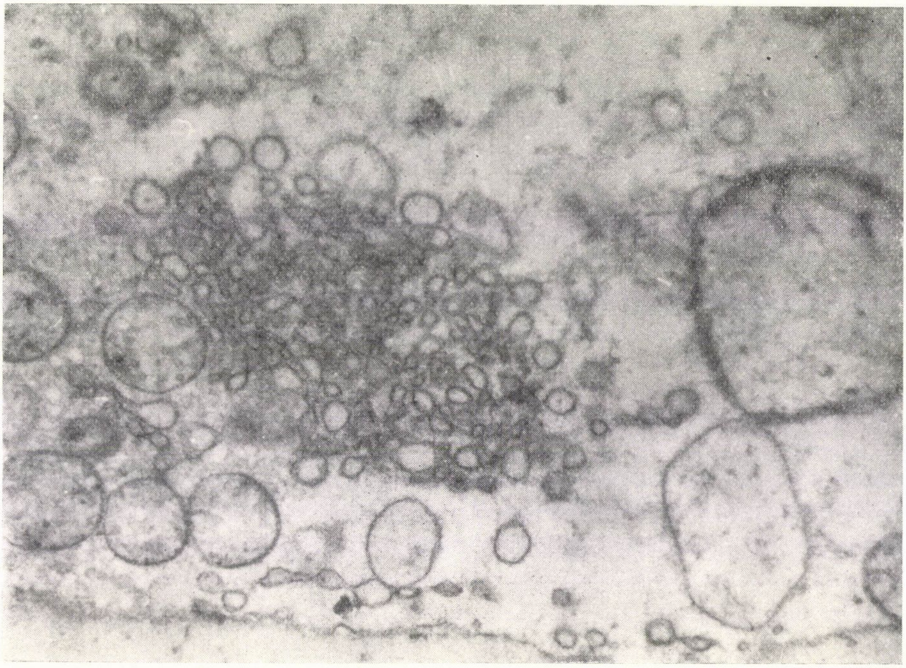

Fig. 4. Cytoplasm part of a meristem cell of *Allium cepa* L. treated with colchicin (explanation in the text). × 13 000

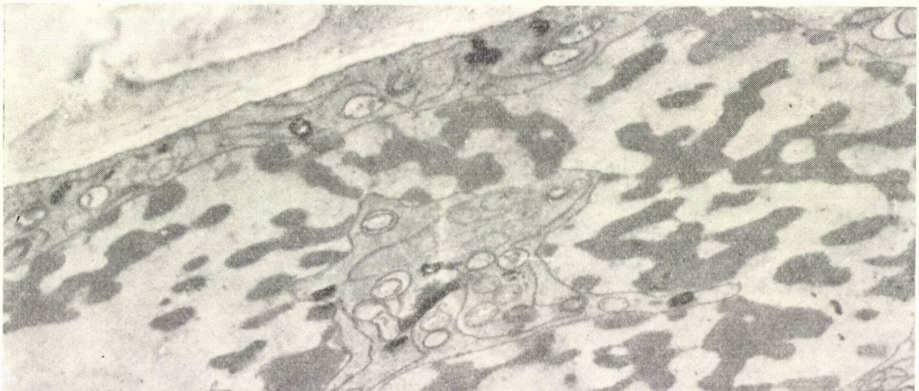

Fig. 5. Part of a meristem cell of *Allium cepa* L. treated with colchicin (explanation in the text). × 4500

can be observed. The next phase is the gradual disintegration of ER into separate vesicula (Fig. 4), and a simultaneous decrease in the number of the Golgi-apparatus. Another essential change can be observed in the nucleus, which is fragmented under the influence of colchicin. This process begins with the membrane of the nucleus becoming introverted here and there and the cytoplasm organelles getting into the creases (Fig. 5). These creases become deeper and deeper, often ramify, and finally the nucleus is divided into two, occasionally even more parts (Fig. 6). Meanwhile an intensive growth and vacuolization of the nucleolus can be observed. From the structural changes described the following conclusions can be drawn. In the apical zones of polarizing or already polarized cells colchicin induces a very intensive temporary protein synthesis as indicated by the high degree of ER accumulation. At the same time the increasing number of mitochondria suggests the growing intensity of oxidative processes. The fragmentation of the nucleus is probably related with the inhibiting effect of colchicin on mitosis, which — as it is known — does not prevent the DNA and the chromosomes from being redoubled. As a final conclusion we can state that an intensive metabolism takes place in the polarizing cells, which is greatly increased for a while by the colchicin.

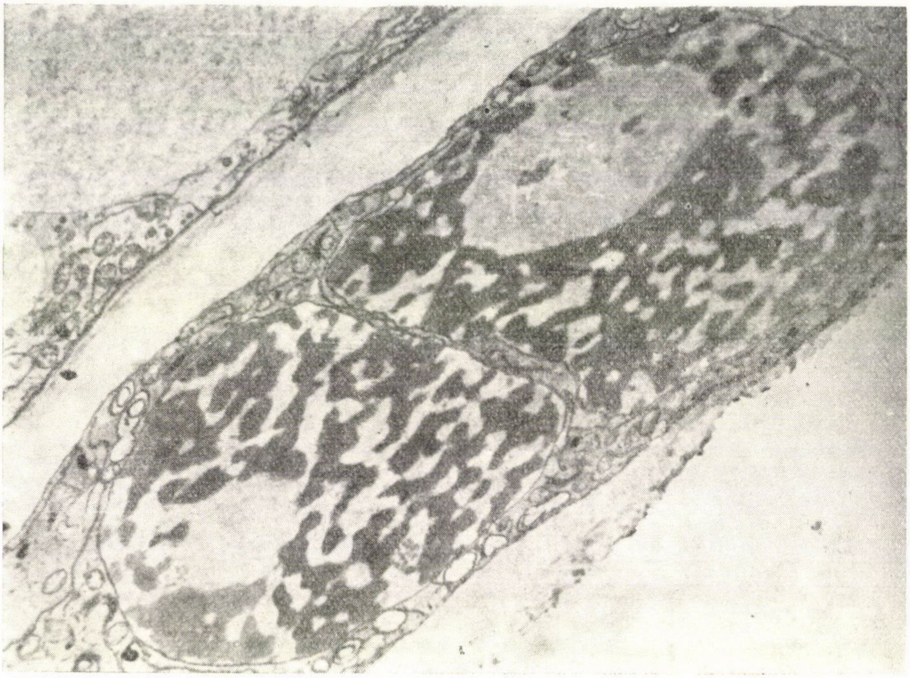

Fig. 6. Meristem cell of *Allium cepa* L. treated with colchicin (explanation in the text). × 3000

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THE GERMINATION PHYSIOLOGY OF TRITICALE

Triticale has lately been successfully grown in Hungary, especially on sandy areas. If attention is paid to harvesting, thrashing and storage seed grains of 85—90 per cent germinative ability can be obtained from the hexaploid hybrid *Triticale* strains produced at Kecskemét (KISS 1968).

According to experiences gained so far the period of dormancy is relatively short due to the high enzyme activity of the grains. Of the numerous factors affecting germination, in addition to the dormancy period, the influence of some salt solutions and of temperature was studied by the author. Grain lots of *Triticale* strains produced and harvested in 1970 (No. 20, No. 64, No. 69, A-1 elite mixture, A-2 elite mixture) were sent by Árpád Kiss (Research Institute of Vegetable Production, Kecskemét) for the experiments. Germination was carried out under laboratory conditions, in Petri-dishes, between filter paper layers kept constantly wet. The required serie of temperatures (5, 10, 15, 20, 25, 30, 35, 40°C) were ensured by the use of refrigerators and biological thermostats. 300 grains per variety with four replications were used in each treatment. In the experiments on salt tolerance (0.5; 1.0; 2.0; 3.0 per cent NaCl, KCl, $CaCl_2$ solutions) and after-ripening the seed-beds were kept at 20°C in darkness, and in the study of after-ripening the two days pre-cooling was carried out in a refrigerator, at a temperature of +4°C.

1. *Study of after-ripening.* At the beginning of October germinating power and germination percentage were in most cases lower than in January or March of the following year. Differences were especially remarkable with the elite mixture strains of A-1 and A-2 which showed 30—40 per cent higher germination ability in January. On the other hand, with the varieties No. 20 and No. 64 the period of after-ripening was short; their grains showed a high germination percentage already in October. With strains A-1 and A-2 the endogenous gibberellin activation induced by pre-cooling (FRANKLAND—WAREING 1962) yielded very good results, especially in the case of A-1, whereas a result of pre-cooling, the germination percentage rose to 93% compared to 62% in the control. The stimulating effect of pre-cooling in increasing the germinating power was found to be similar in the other varieties (Table 1).

2. *The cardinal points of germination temperature.* At 5°C germination only began on the 9th or 10th day, at 10°C on the 6th day, while at 15°C and over as soon as on the 3rd day. At 0°C not a single grain of *Triticale,* while at 40°C only 1—2 per cent germinated. The grains of strain A-1 showed a remarkably good germination percentage already at 10°C, while the variety No. 64 germinated to 3 per cent even at 40°C. The optimum germination temperature

Table 1

Effect of pre-cooling on the germinating power and germination percentage of Triticale grains
(Tápiószele, 1970—71)

Variety	Water content in grains Oct. 1970 %		Number of germs on the 2nd day of germination (Oct. 1970)	3rd	4th	Germination percentage (Oct. 1970)	Germination percentage (Jan. 1971)
No. 20	9.4		67	13	6	86	86
		pre-cooling	81	0	0	81	
No. 64	8.6		42	33	13	88	87
		pre-cooling	82	4	2	88	
No. 69	9.1		27	22	14	63	71
		pre-cooling	51	9	5	65	
A-1	! 7.7		29	7	26	62	94
		pre-cooling	84	7	2	! 93	
A-2	! 7.7		13	11	27	51	89
		pre-cooling	71	10	2	! 83	
Mean						70	85
						82	

Table 2

Germination percentage of Triticale varieties at different temperatures
(Tápiószele, 1971)

Variety	Range of temperature °C 5	10	15	20	25	30	35	40
No. 20	62	79	84	*86*	84	65	64	0
No. 64	70	65	66	83	*88*	86	69	3
No. 69	40	51	54	*71*	64	59	38	0
A-1	83	94 !	94	*94*	87	70	69	0
A-2	65	88 !	88	*89*	74	54	51	0
Mean	64	75	77	*85*	79	67	58	1

was around 20°C with most varieties except No. 64 which showed the highest germination percentage at 25°C (Table 2).

3. *Effect of salt solutions.* Examinations were performed on two occasion: at the beginning of the period of after-ripening (early October, 1970) and nearly half a year later (at the end of March 1971). The grains of strains with a longer after-ripening period (A-1 and A-2) were more sensitive at the beginning of the period than those of varieties with a shorter after-ripening period (No. 20, No. 64, No. 69). NaCl reduced the germination percentage at as low a concentration as 0.5 per cent, and at 2.0 per cent even showed a toxic effect. Germi-

Table 3

Effect of salt solutions on germination in Triticale
(Tápiószele, October 1970)

Variety	Control	Germination percentage after treatments with								
		NaCl			KCl			$CaCl_2$		
		0.5	1.0	2.0	0.5	1.0	2.0	0.5	1.0	2.0
		per cent solutions								
No. 20	86	84	46	1	81	59	8	69	77	45
No. 64	88	87	34	6	79	67	9	91 !	77	38
No. 69	63	63	30	1	68 !	47	10	57	41	12
A-1	62	25	12	0	37	36	5	48	35	4
A-2	51	24	10	1	36	28	7	47	25	3
Mean	70	57	26	2	60	47	8	62	51	20

Table 4

Effect of salt solutions on germination in Triticale
(Tápiószele, March 1971)

Variety	Control	Germination percentage after treatments with											
		NaCl				KCl				$CaCl_2$			
		0.5	1.0	2.0	3.0	0.5	1.0	2.0	3.0	0.5	1.0	2.0	3.0
		per cent solutions											
No. 20	90	80	84	44	10	86	76	85	51	77	79	70	69
No. 64	94	93	76	50	7	83	82	68	63	92	78	76	68
No. 69	70	69	53	19	2	71	49	42	24	68	60	54	36
A-1	96	90	83	36	3	94	81	52	28	92	91	81	67
A-2	89	78	47	19	2	72	69	23	14	86	83	75	42
Mean	88	82	69	34	5	81	72	54	36	83	78	71	58

nation was decreased in the lowest degree by the $CaCl_2$ solutions; in the variety No. 64 the germination percentage even increased from 88 to 91 per cent under the influence of a 0.5 per cent $CaCl_2$ treatment; while the 0.5 per cent KCl treatment increased the germination percentage of the same variety from 63 to 68 per cent (Table 3).

On the second occasion (at the end of March 1971) the varieties displayed less susceptibility; germination did not fall to zero even with 3 per cent salt solutions used. The strain A-1, with a high germinative ability as well as grains of strains No. 64 and No. 20 germinated even when treated with 3 per cent NaCl (germination percentage was 3—10) (Table 4).

It is known that rye grains are highly salt tolerant while wheat grains are more sensitive (MÁNDY—PÁL 1960). It was interesting to discover that the *Triticale* grains were more sensitive to higher concentrations (2—3 per cent) of salt solution than the simultaneously germinated rye grains. The salt tolerance of Triticale grains is about the same level as that of wheat and oat grains (Table 5).

In the germination physiological study of *Triticale* the rate of ripening of grains was found to be different in the various strains. At the beginning of the after-ripening period pre-cooling had a highly favourable effect on the grains of strains A-1 and A-2. Seed grains of these varieties are more sensitive in this period even when treated with salt solutions. In the course of the after-ripening process, about half a year later, grains of *Triticale* varieties germinated well; they showed a low per cent germination even when treated with the highly inhibiting 3 per cent solution of NaCl. Compared to rye *Triticale* has a lower salt tolerance, similar to that of wheat and oat grains.

Table 5

Effect of NaCl solution on the germination of grains in some cereals
(Means of 5 varieties per each)
(Tápiószele, 1971)

Corn species	Control	Germination percentage after treatments with	
		2.0	3.0
		% NaCl solutions	
Wheat	95	67	7
Rye	74	73	63
Triticale	88	34	5
Oat	92	62	2

When studying the cardinal points of germination temperature in *Triticale* varieties we found 20°C to be about the optimum, 35°C the maximum (40°C with only one of the varieties examined) and 5°C to be the minimum temperature.

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EFFICACY OF FUNGICIDES IN CONTROLLING COLLAR ROT OF GROUNDNUT (ARACHIS HYPOGEA L.)

There are conflicting reports regarding the efficacy of organo-mercurials in controlling *Aspergillus niger* van Tieghem, causing collar rot of groundnut. Morwood (1945) reported that Agrosan G. N. and Ceresan (toly mercury acetate and ethyl mercury chloride respectively are effective in controlling *A. niger*. Later, Morwood (1953) and Purss (1960) observed that the post-emergence loss of plants was higher in seeds treated with organo-mercurials than in seeds treated with other fungicides. Nema *et al.* (1955) tested various organo-mercurials and observed that all the materials reduced the pre- and post-emergence death of groundnut plants as compared to the control. Studies were, therefore, undertaken to test the efficacy of some newly developed organo-mercurials along with other fungicides, for inhibiting the growth of the pathogen *in vitro* and controlling the disease and the results are presented here.

The bioassay was carried out with the method described by Sahni—Singh (1967). A spore suspension from a ten-day-old single spore pathogenic culture of *A. niger* was prepared, in distilled water and flooded in petri dishes having 2% potato dextrose agar (PDA). Seeds of groundnut (variety local) were treated thoroughly with seven concentrations (0.1, 0.2, 0.3, 0.4, 0.5, 0.75 and 1.0%) of each of the fungicides (Agrosan G. N., Ceresan wet, Fertix 85, Harvasan, Seed Dresser 6334, Seed Dresser 6335, Brassicol and Thiram). Three seeds from each treatment in five replications and the same number of untreated controls, were placed on the surface of the inoculated PDA in petri dishes at equal distances. The average size of the inhibition zone produced around germinated and ungerminated seeds was recorded on the sixth day.

To study the efficacy of fungicides in controlling the disease, seeds were inoculated with the spores of *A. niger* from a 20 day old culture and treated separately with fungicides at concentrations shown in Table 2. Surface sterilized, unsterilized and inoculated seeds without fungicidal treatment were used as controls. Four seeds from each treatment were sown in each of twenty five 12 inch pots having sterilized field soil. Observations were recorded on the number of seeds germinated, length of shoot, number of diseased plants on the 10th, 15th and 20th day of sowing.

Data presented in Table 1 show that Seed Dresser 6335 was the best at producing larger inhibition zones at all concentrations, followed by Seed Dresser 6334 and Fertix 85. Brassicol

Table 1

Growth inhibition of A. niger by fungicides

Fungicides	Average diameter of inhibition zones							
	Concentration of fungicides in %							
	0.0	0.1	0.2	0.3	0.4	0.5	0.75	1.0
Agrosan G. N. (Toly mercury acetate)	Nil	1.45	1.60	1.80	1.85	2.30	2.50	2.80
Ceresan wet (Ethyl mercury chloride)	Nil	1.10	1.50	1.90	2.20	2.35	2.40	2.65
Fertix 85 (3% mercury as organo-mercury compound)	Nil	1.60	1.80	2.40	2.60	2.85	3.00	3.20
Harvasan (Organo-mercury compound)	Nil	1.30	1.60	1.90	2.15	2.30	2.40	2.70
*Seed Dresser 6334 (3% mercury as organo-mercury compound)	Nil	2.10	2.40	2.70	3.10	3.40	3.70	4.00
*Seed Dresser 6335 (3% mercury as organo-mercury compound)	Nil	2.30	2.90	3.20	3.40	3.80	4.00	4.20
Brassicol (Pentachloro nitrobenzene)	Nil	Nil	0.90	1.20	1.65	1.80	2.00	2.40
Thiram (Tetramethyl thiuram disulphide)	Nil	1.30	1.40	1.70	2.10	2.20	2.35	2.60

* Coded product of M/S Sandoz Ltd., Basle, Switzerland.

which is a good fungicide for various soil borne fungi, gave the poorest results. It was observed that in the control *A niger* grew all round the seeds and inhibited their germination. Seeds germinated well in all the treatments except in those of the higher concentrations (0.2% and above) of Seed Dresser 6335. After initiation of germination, further growth stopped, indicating that the fungicide is toxic at these concentrations.

Table 2

Efficacy of fungicides in controlling Aspergillus collar rot of Groundnut

Treatments	Germination	On 10th day		On 15th day		On 20th day		% Disease Control
		A	B	A	B	A	B	
Agrosan G. N. .4% .... (Toly mercury acetate)	98	11.42	Nil	16.50	4	17.97	4	92.49
Ceresan wet. 2% .... (Ethyl mercury chloride)	94	11.37	Nil	15.43	2	15.80	5	90.56
Fertix 85 3% .... (3% mercury as organo-mercury compound)	100	11.64	Nil	16.57	Nil	19.31	Nil	100.00
Harvasan 3% .... (Organo-mercury compound)	95	11.54	3	15.27	4	15.75	7	86.79
*Seed Dresser 6334 .3% .... (3% mercury as organo-mercury compound)	97	11.80	Nil	16.57	Nil	18.71	Nil	100.00
*Seed Dresser 6335 .2% .... (3% mercury as organo-mercury compound)	100	12.03	Nil	17.22	Nil	19.42	Nil	100.00
Brassicol 5% .... (Pentachloronitro benzene)	92	10.13	3	14.80	5	15.52	8	84.92
Thiram 3% .... (Tetra methyl thiuram disulphide)	98	11.33	Nil	15.97	2	16.88	2	96.22
Sterilized	98	10.88	2	14.41	5	15.70	8	84.92
Untreated	89	8.44	5	11.21	7	12.54	12	77.35
Inoculated	70	4.33	12	6.86	36	7.77		50.003

* Coded products of M/s. Sandoz Ltd., Basle, Switzerland
A = Average length of shoot (cm)
B = Number of seedlings rotted

Results presented in Table 2 show that inoculated and unsterilized and not inoculated seeds had a germination percentage of 70 and 89 respectively whereas seeds treated with fungicides or surface sterilized had a germination percentage between 92—100. Fertix 85, and Seed Dresser 6335 treated seeds had a hundred per cent germination. Fertix 85, Seed Dresser 6335 and Seed Dresser 6334 could control the disease up to 100% followed by Thiram and Agrosan G.N. which could control it up to 96.22 and 92.49% respectively. It was interesting to note that the length of the shoots was retarded in the treatment inoculated with the pathogen while same increased in all other treatments. Maximum shoot-length was recorded in the case of Seed Dresser 6335 followed by Fertix 85, Seed Dresser 6334 and Thiram.

Inhibition zones of maximum size were formed by Seed Dresser 6335 followed by Seed Dresser 6334 and Fertix 85. However, it was observed that Seed Dresser 6335 is phytotoxic at a concentration of 0.2% and higher. The comparative efficacy of these fungicides in controlling the disease was found to be better followed by Thiram and Agrosan G. N. Thus it will be seen that these mercurials (Seed Dresser 6335, Seed Dresser 6334, Fertix 85) and Thiram are better as compared to other mercurials like Agrosan G.N., Ceresan wet and Harvasan for controlling the disease. MORWOOD (1945) and NEMA *et al.* (1955) reported that organo-mercurials could control the disease but later MORWOOD (1953) observed that the post-emergence loss of plants appeared to be less in seeds treated with Thiram. PURSS (1960) also reported that organo-mercurials were effective in preventing pre-emergence rotting, however they were associated with a significantly higher incidence of the disease than that in the case of treatments with Captan and Thiram. GIBSON (1953) reported that organo-mercurials produced a substantial increase of the disease attributing this to the reason that certain isolates of *A. niger* were mercury tolerant and concluded that this effect of the organo-mercurials was due to their selective action upon the fungal flora of the soil region of the treated seeds operating to the advantage of *A. niger*. In the present studies too Thiram was found to be better than Agrosan G. N., Ceresan wet and Harvasan but inferior to Seed Dresser 6335, Seed Dresser 6334 and Fertix 85 indicating that this isolate is not mercury tolerant but these mercurials have a better fungicidal action. SHARMA—AGNIHOTRI (1969) observed that the mycoflora associated with the groundnut seeds could effectively be controlled by Seed Dresser 6335 and Seed Dresser 6334 and that *A. niger* and species of *Fusarium* and *Sclerotium* could not completely be inhibited by Agrosan G.N., Brassicol, Harvasan, Ceresan wet and Thiram.

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LENTIL IREGI CIRMOS

Taxonomical place: *Lens culinaris* MEDIK. convar. *microsperma* BAR. provar. *intermedia* (BAR.) MY conc. *subnummularia* BAR.

Origin: Selected from a local variety of French origin.

Beginning of breeding: 1958, Iregszemcse

Breeder: Béla Kiss dr. and László Mészáros, Iregszemcse

State qualification: Provisionally certified improved variety, 1966.

General characterization: Productive, high yielding, uniform early variety with light brown, spotted seeds.

Morphological description:

Root system: strong main root penetrating to a depth of 60—80 cm (MÁNDY 1971)

Shoot system: mostly low, bushy plants of an average height of 35—37 cm, with an inclination to lodging (PAPP 1971). Surface slightly hairy.

Foliage: dense; lower leaves have seven, upper leaves seven or eight pairs of leaflets; the rachis is often terminated by an odd-branched tendril. Leaves are of a medium green colour. Leaf storeys in the middle zone have dentate edged stipulae of about 4 mm length at their base.

Flowers: racemose inflorescence with 2—4 flowers. The flowers are generally small and white with intensive violet veins. The standard is 5.5—5.8. mm long and 5—6 mm broad (SOMORJAI 1970, PAPP 1971).

Pod: 8—15 mm long and 6—7 mm broad, prismatic, and mostly contains two seeds when mature.

Seed: of 3—5 mm diameter, flat, disc-shaped; multicoloured, with a drab or light greenish-brown ground colour and greenish-dark blue streaks. Hypocotyl yellow. Thousand-grain-weight 25—35 g (SOMORJAI 1970).

Biological characters:

Germination: minimum temperature required 3°C, optimum 20°C, maximum 40°C. Germination power very good, at an optimum temperature seeds germinate abundantly as early as on the second day (PAPP 1971). Sprouting 12—20 days.

Vegetation period: early ripening variety with a short vegetation period (100—110 days); flowering from the end of May.

Development: rapid and vigorous; ripening uniform and quick.

Resistance to diseases: fairly good.

Farm technology requirements:

Sowing: early in April, at a depth of 5 cm.

Soil requirements: high yielding in fertile soils.

Productivity: average seed yield 7.8 q/cad.yoke (1 cad.yoke = 5754.56 m^2). fluctuation 5.3—8.6 q/cad.yoke. Straw weight somewhat more than seed weight (ratio = 1 : 1.2). Well adaptable to extreme weather conditions. Quality of seed is good, taste better than in other streaked varieties.

Area of cultivation: in soils suitable for lentil production succesfully grown everywhere in Hungary.

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Prepared at the University of Agrarian Sciences, Dept. of Botany, Debrecen.

GY. MÁNDY

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LECTIONES

PROBLEMS OF IMPROVEMENT IN CATTLE*

Improvement in cattle is both objectively and subjectively aggravated.

Along with the increasing productivity level the genetic variance of the characteristics becomes smaller and the breeding progress, pro unit of time, less considerable. The more consequent the breeding within a population, the sooner and more intensively the formation of a plateau appears. The result of such an expression of characteristics is the best precondition for new and more advanced breeding purposes by using combinations of various populations selected in a similar direction. In plant breeding, considerable results have been achieved with this method of transgressive breeding.

Objective barriers to new breeding projects. The methods of animal breeding when compared to those of plant breeding are limited. At the same time the biological possibilities are in general — hardly used in comparison to plant breeding, see Fig. 1.

The systematic use of inbreeding is avoided and similarly the great variety of crossbreeding methods are also not methodically applied in modern breeding work. The reasons for the differentiated use of the possible breeding methods in commercial plants and animals are to be searched in the objects themselves, first of all in the value of the individuals, their reproductive performance and in the generation interval respectively. Owing to these one has to account with considerably higher economic investments in large animal breeding than in plant breeding. The new cattle breeding projects cannot be realized independently of the breeding enterprises and the leading breeding organizations of the respective country. These experiments with high risks are additionally linked with a considerable interval between the investment and the amortization.

Subjective barriers for the new breeding projects. The problem originates not only from the too far-set purposes in such projects. It already begins with the general aversion to all kinds of cross-breeding in cattle. Finally the facts of the case are reflected in the legislation and the bye-laws of the breeding organizations by which even the well-established projects are at least hindered if not completely banished. This very often happens with the argumentation, that breeding has to be protected against undesirable segregation. Already Wright and above all Robertson, after analysing extreme breed-crosses of cattle, have very definitely disproved the justification of fear from segregation of production characteristics. Crossbreeds within dairy cattle, owing to the decreased reproductive capacity can only have the most important aim of utilization of additive gene effects. These must be used wherever they are offered, independently of whether the mating is a conventional cross-breeding or not. Should, however, in such cross-breeds, non-additive gene effects also occur, they never stand in the limelight in the case of cattle.

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* The paper was presented on the International Meeting of Animal Breeding held in Budapest.

As far as the setting of objectives in concerned, there is no basic difference between pure breeding and cross-breeding in cattle. Both methods take into consideration the parent-offspring similarity, on which Fewson's suggestion regarding the repartition of the breeding methods is based. A qualitative evaluation of the two methods is unjustified. Both of them are complementary to each other and can be developed only by being exchanged in chronological succession. Which of the two methods should be favoured under given conditions can be decided exclusively on the basis of the genetic production parameters of the respective populations and of the future economic-technological demands of the respective country.

Moreover it seems that a very probable superiority of new populations can often be the greatest barrier to their expansion. In many leading animal breeding countries the examination and the introduction of foreign populations is a priori prohibited or limited. At this point, there are also various argumentations for the high quality and multilateral perfection of the indigenous breeds within the countries. In periodicals of breeding associations this is well understandable, in technical journals, however, it should be avoided, for it supports self-satisfaction and similarly hinders the inbreeding.

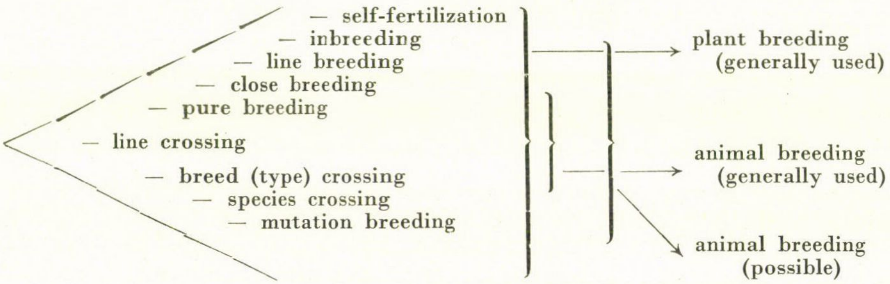

Fig. 1. Breeding methods that can be used in plant and animal breeding

The 6th International Congress on Animal Breeding in 1952 at Copenhagen published the obligatory final explanations of experts on the results of the discussions. As regards breeding methods the explanation says, inter alia; "Cross-breeding as a breeding method in the form of grading is entirely reasonable, when speaking about the improvement of primitive breeds or about that of highly developed breeds in which certain characteristics, connected with conformation and production still need some improvement". These formulations satisfied the general requirements of the breeding practice as well as the demands of animal production nearly two decades ago. Thus the directives were valid for the tasks of research and breeding practice of the 50's and 60's.

The production technology of the future, however, is already setting up requirements regarding animal populations which can no longer be reliably solved by using only selection within the breed standards of each country. This, however, would not be a reason for changing the conception until no basic genetic and breeding technological solutions are to be found. If opposite to this, even today, the cross-breeding methods are "fully accepted" only for improving "primitive breeds" and such improved ones in which "certain characteristics as regards conformation and production" are still to be improved, the highest European authority of animal breeding has determined restrictions for the breeding possibilities which no longer conform with the present status of animal genetics, breeding research, and breeding techniques.

Besides economic considerations, the important preconditions for the solution of new projects in cattle breeding are

— the general acknowledgement of their genetic breeding reality and

— the support of such projects without reservation through breeding practice, i.e. breeding organizations possibly on the basis of an international cooperation.

Both of the preconditions are underdeveloped as yet. The European Association for Animal Production could also decisively contribute here to both points.

The genetic-breeding grounds that are available for new breeding projects are to be judged favourably at an international level.

Table 1

The relative realization of characteristics of dairy and dual purpose breeds

Breed	Characteristic				
	Milk kg	Fat %	Udder	Fattening perform.	Slaughter-value
Jersey	+	++	++	—	—
Holstein-Friesian	++	—	+	++	—
British-Friesian	++	±	+	++	+
European-Friesian	+	±	±	++	+
European-Fleckvieh	—	+	—	++	++

The animal breeding organizations of the leading animal breeding countries of the world have developed during many decades of systematic breeding work, breeds with excellent abilities for special production characteristics and all these have been genetically fixed by consequent cattle breeding. With regard to the characteristics of economic significance, a tandem-selection with differentiated selection pressure amongst populations has taken place. As a result of this, there is a well prepared basic material available for improvement and combinative cross-breeding according to the special requirements in the respective countries and territories, see in Table 1.

This "gene-pool" meets the requirements that are necessary for far-reaching breeding results and it is required immediately afterwards, as a source for further increase of production to be opened systematically.

Unfortunately the data of the breed-characteristics cannot be further specified as internationally comparable breed tests are lacking. These also should be carried out with the support of the EAAP in which strong measures, supporting breeding work can be seen.

Also in cattle breeding one has to work on these tasks of improvement with the same seriousness and care as on the systematic development of the available highly productive breeds. Many experimental results of combinative cross-breeding speak in favour of a good amortization of these costs: Wright, Schmidt, Cole and Johansson, Fohrman et al., Arzumanjan, Merkujewa, Horn, Lebedew and Gorjasin, Schönmuth, Lenschow and Tilsch, Leuthold, Hanschmann, Jasiorowski, Ernst, Dohy and others.

Our own performance analysis carried on since 1961 was performed with the genotypes German Black and White (DSR), Danish-Jersey, British-Friesian and their combinations: F_1, R_1 and with the three-breed construction with 25% D-Jersey gene ratio. The following characteristics were submitted to analysis: raising performance, fattening performance, slaughter-value, early maturity, milk production and milkability.

Keeping in mind that an up-to-date technology requires high butterfat and milk protein production and under European conditions (intensity, housing of animals in winter time) the same dairy cow population must also be the basis of the rapidly increasing need for beef, it seems reasonable to aim at the following gene-combinations

50% Friesian
25% D-Jersey
25% DSR (German Black and White)

Genetically the following mating plan is offered:

F_0 DSR ♀× Danish-Jersey ♂
F_1 × British or Holstein Friesian
"F_2" mating among themselves

If the slaughter value had a greater economic importance than at present Graevert, Rittler, Kreusslich, then later a certain part of the population could be mated to a male line, selected for beef production. The actual superiority of the so-called beef breeds would not, however, satisfy the requirements as shown by Weniger and Engelke.

The mentioned construction compared to a German Black and White population offers the following improvements: Early maturity, butterfat production, milk concentration and udder quality. The disadvantageous effect of the Danish Jersey cattle as regards beef and raising performance will be compensated by the respective superiority of the Friesians.

On the basis of our extensive cross-breeding experiments we think we have to come to the conclusion that many productive breeds offer combinations from which populations with higher performance superiority can be developed. For example a considerably more rational absolute as well as relative energy-utilization and performance superiority is shown by the F_1 population deriving from the cross-breeding: Danish-Jersey ♂× German Black and White than by the German Black and White breed, see Table 2. The reserves hidden in the breeding work are shown e.g. by the butterfat production (energy performance) pro kg live weight. The F_1 animals have overtaken the German Black and White individuals by 42% unless the general production characteristics such as vitality, fertility etc. had been affected.

Table 2

Absolute and relative performance in FCM and feed utilization of F_1 cows deriving from the mating: German Black and White (DSR) ♀× Danish-Jersey compared to DSR (first lactation)

	F_1 (n = 12)		DSR (n = 11)			$\bar{x}$ relat. DSR = 100
	$\bar{x}$	s %	$\bar{x}$	s %	p %	
Total FCM kg	3751	14.23	3187	13.18	0.96	117.7
Weight gain ± kg	+41.7	97.07	66.6	53.85	13.00	62.5
Corrected FCM, kg	4168	16.54	3853	15.78	25.00	108.2
FCM pro kg live weight kg	8.2	18.29	6.36	16.04	0.22	128.9
Butterfat pro kg live weight kg	0.371	18.33	0.261	17.62	0.10	142.2
g StE for kg corrected FCM	428	7.94	462	7.66	2.9	92.6

The actual economic interest of the countries sometimes makes possible or claims specific conceptions, in part, even contradictory ones. The status of the breeding technique makes it possible — at least on theoretical and economic grounds — without using the approved methods of cattle breeding to create new productive populations which are first of all predestinated for industrial-like production methods.

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POSSIBILITIES OF FURTHER IMPROVEMENTS IN SWINE PRODUCTION*

Almost all characteristics which are of importance regarding the economy of swine production and the quality of the products are depending both on genetic factors and the environment. A future improvement in swine production is therefore depending on improvement of the heritable characteristics of the individuals and on the environmental factors under which the animals live.

Economic factors of the swine production

Fertility, daily weight-gain and feed utilization are characteristics, which have a genetic basis. Since, however, the heritability estimates are rather low (0.1—0.2), in my opinion one should have to concentrate on improving the environmental conditions (feeding, care and housing conditions) under which the pigs will be kept.

These characteristics with lower heritability, can also be improved by means of commercial crossing between breeds, or lines within one breed (hybridization).

The constitution of the animals is the inherited ability to resist the environmental influences diverging from the optimum. This definition can be supported among others by the following experimental results. Under the same environmental conditions on the Danish performance testing stations less English Large White pigs had to be culled as of Danish Landrace ones (Clausen *et al.* 1954). Einarsson (1968) observed that in artificial insemination the Large White boars served considerably longer than those of the Landrace breed. During the first weeks after the birth the haemoglobin level in the piglets' blood has a great importance as regards resistance against various diseases.

Under similar environmental conditions differences were observed among sow families of the Danish Landrace breed (Jespersen *et al.* 1939) Paul Jensen (1964) found a heritability of 0.17 for the haemoglobin level in the blood of piglets at birth.

The frequency for pale soft exudative meat (PSF) is higher in the Belgian Pietrain pigs than in those of the Danish Landrace (Clausen *et al.* 1961) and in the USA it is higher in the Poland-China breed than in the Chester White and Duroc breed (Ludvigsen 1968). The meat quality and as a consequence of this the occurrence of pale watery meat is partly determined by additive gene effect in the Danish Landrace ($h^2 = 0.31$, Jonsson 1966).

Thielscher compared the electro-cardiogrammes of animals from two different breeds (Pietrain and German Landrace), which were submitted to a controlled stress (running) in a special equipment and found a significant difference between the two breeds. Atrophic rhinitis, the serious alteration of the nose is partly implicated with an additive gene effect. Jonsson (1966) found that the heritability estimate of this trait in Danish Landrace pigs examined in the Danish fattening performance testing stations was as much as 0.18.

Indirectly a number of anatomic traits have influence on resistance and the duration of use. A greater body length raises higher requirements toward the strength of the back and legs especially toward that of the hock joint. Too small hoofs may reduce the strength of the limbs.

Along with the disappearance of the bristles and the decrease of the back fat thickness the pigs become more sensitive to the unpreferable housing conditions (draught, fluctuation of temperature).

In my opinion along with the future increase of the selection intensity of breeding animals aimed at the improvement of certain important characteristics, the conformation of the pigs has to be taken into consideration to a greater extent. New methods have to be

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* The paper was presented on the session of the Hungarian Academy of Science held on 13th May, 1971.

evolved for the more exact evaluation of the constitution of the individuals ment for breeding. This is particularly important in the case performance testing is practised.

As the heritability estimates of the characteristics, being of importance in evaluating the constitution of pigs, are low, one can count with a heterotic effect in this field also.

Along with the centralization within swine production into less, but larger units the problem of keeping animals free from serious epidemic diseases becomes highly important. I am referring here to diseases such as ensootic pneumonia, pig dysentery and infectious enteritides of newborn piglets.

The establishment of among others "Pig Health Central Associations" (Great Britain), "Beratungs- und Gesundheitsdienst in der Schweineproduktion" (Switzerland, Denmark) and "Helskontrollen" (Sweden) will be necessary also in many other countries in the future if an increase in productivity should be achieved.

At the same time one must pay special attention to the set-up of SPF (specific-pathogen-free) pig stocks that are already available in the USA, Great Britain, Switzerland, Germany, Austria and now also in Denmark.

There is no doubt, however, that a more intensive research on the foetal mortality (foetal atrophy) and the mortality of piglets between birth and weaning might considerably decrease the expenses of production. One may therefore look forward with great expectation to the set-up of experimental work in the field of the feeding of pregnant and nursing sows as well as piglets.

The birth of piglets by means of hysterectomy (Caesarean section) or the early weaning of piglets at 4, 12 or 21 days of age can probably be carried out only in large-scale-farms and owing to possible infections, only in closed herds or in such ones, where artificial insemination is practised.

The question of housing conditions also belongs to the problems of production economy. The development of labour-saving technical equipments, along with the increasing labour costs, is reasonable. While, aiming at a high meat production with animals from highly improved breeds, increased requirements of climatization of pig houses and sanitary conditions are indispensable. It means ventilation and in certain countries also heating as well as hygienic measures in order to keep health disorders at a low level and to establish the possibilities for the production of SPF piglets.

If the development of feeding, care and the establishment of more convenient housing conditions do not keep step with the breeding work, a dangerous discrepancy may occur between the elite herds and its achievements on the one, and the production and its economy on the other hand.

Slaughter value

In such countries, where the quality of the slaughtered pigs has not a great importance generally all activities can be restricted to the improvement, of the economic factors of the production if however one is interested in increasing carcase quality then much higher requirements are set both to the pig breeders and to the pig producer. A progress depends not only on the efficiency of selection of the animals but also on certain optimal environmental conditions, under which the pigs have to grow up. In this respect the feeding has a particular importance.

There are three fundamental laws for the meat and fat formation in pigs (CLAUSEN 1959):

1. No pig can realize its genetically determined muscle flesh producing capacity if the food does not contain enough protein of high biological value.

2. No pig can be forced, by means of feeding with extremely high protein containing feeds to produce more muscle flesh than determined genetically.

3. If the daily requirement for maintenance and muscle flesh production is covered, all the feed consumed in excess must be used for fat production. This occurs rather early in case of pigs with poor muscles.

The meaning of these laws is that in the feeding of pigs, besides the mineral and vitamin requirement so much protein has to be provided, that the genetically determined meat producing capacity is completely utilized without the manifestation of a luxury consumption of the scanty and expensive protein feeds. At the same time one has to restrict the calory supply to such an extent, that fat production is kept as low as possible.

These rules form the basis for the restricted feeding that in my opinion is generally valid until a new pig type is developed, which is able to produce so much meat daily, that it can be fed ad libitum without any risk.

Table 1

Lysin requirement of growing meat type pigs

	Gramme	Lysin	Per day	Per pig	
Body weight kg	20	30	50	70	90
According to Clausen	8.1	11.6	14.7	17.6	19.7
According to Poppe and Wiesemüller	9.0	12.0	15.0	16.4	17.3

There are now automatic feeders which measure out the feed according to capacity with high accuracy so that the restricted feeding does not need the expensive labour for weighing feed rations which is necessary in each feeding system.

Danish experiments aimed at the reduced calory content of the feed used in ad libitum feeding, by means of an increased crude fibre content. This however did not produce any reliable result partly because crude fibre is poorly utilized by the pig and partly as the high crude fibre content of the feed has an undesirable influence on the solidity of the back-fat (CLAUSEN 1959).

Law No. 1 regarding meat production of the pig involves the concept "enough amount of protein". This concept says that *if the possible highest meat production is wanted* then the actual protein level of the ration has to be over the physiological protein norm the average of which has been established in female and castrated animals. The average norms calculated for female animals are also inadequate as the food must contain so much protein that the needs of animals possessing the best genetic basis for meat production can also be satisfied.

As it is generally known, it is also insufficient that the food contains adequate amount of crude protein or digestible pure protein. Pigs fed with the same amount of digestible pure protein in the course of their growing period may show quite different meat/bone ratio at slaughter. Certain amino acid level in the feed is essential for meat production. The needs of lysin and other sulphur containing amino acids (methionin and cystin) have been experimentally determined among others by CLAUSEN (1963) and POPPE—WIESEMÜLLER (1968). The experimental results regarding the lysin requirement are indicated in Table 1.

The values for 20—50 kg live weight, are in good agreement. The Danish experiments show higher requirement for the 50—90 kg weight classes that can be explained, perhaps, by the fact that the Danish experimental animals (Danish Landrace) possessed a genetic basis for a higher meat production at least in these classes, than those used in the Rostock (Germany) experiments.

It is an important problem for our protein starving world whether the protein requirement which is absolutely necessary for the meat production of pigs has to be covered by the amino acid content of natural foodstuffs or by industrially, i.e. microbially or synthetically produced amino acids. This question will be discussed later more profoundy.

The laws of meat production of pigs and their importance, regarding the quality of the meat have also been elucidated by the works of STAUND (1963, 1968).

It is a generally accepted conception that the total number of muscle fibres in pigs cannot be increased after birth. In feeding experiments it was absolutely impossible to modify the number of muscle fibres in the *musculus longissimus dorsi* either by the intensity of feeding (poor, medium, normal, intensive, highly intensive) or by changing the protein content in the feed (low, medium or high). The genetic disposition for the total number of the muscle fibres and their development are decisive for the development of a large back musculature. The feeding is of importance only during growth of the muscle fibres and in consequence of this in the meat ratio as well.

Table 2

The effect of feeding-intensity on the meat production

Intensity of the feeding	Poor	Medium	Normal	Intensive	Highly intensive
Diameter of muscle fibres (μ)	72.5	74.7	70.8	64.0	63.9
Surface of m long. dorsi (cm^2)	32.2	31.1	29.6	28.1	27.5
Meat % in the halved carcases	58.1	56.0	54.1	51.8	51.8

Table 3

The effect of protein content of the feedstuff on the meat production.

Protein content of the food stuff	Low	Medium (normal)	High
Diameter of muscle fibres (μ)	66.7	70.3	71.0
Surface of m long. dorsi (cm^2)	27.0	31.1	31.0
Meat in % of the halved carcases	51.3	55.6	56.2

In the case of highly intensive feeding, the pigs reach the slaughter weight earlier, therefore the muscle fibres cannot develop as wide (shorter time) as in the case of less intensive feeding wherefore the meat ration remains too small at a given slaughter weight.

Owing to too small protein content in the feed the muscle fibres are less developed (or develop, slower) and as a consequence of this the meat ratio at slaughtering will be too low. An overfeeding with protein increases the diameter of the fibres and the meat ratio to a minimal degree.

Our investigations on the muscle fibres have shown, that the heritability of the diameter of the muscle fibres is as much as 0.2 (in females $h^2 = 0.31 \pm 0.23$; in castrated animals $h^2 = 0.17 \pm 0.24$) while the heritability of the total number of the fibres in the m. long. dorsi is 0.8 (in female animals $h^2 = 0.88 \pm 0.29$; in castrated ones $h^2 = 0.66 \pm 0.29$). The meat with a great number of thin muscle fibres per mm^2 is softer than that with a few number of rough ones.

It will be a future breeding task to take aim both at the m. long. dorsi with larger surface and at more fibres per surface unit as in this way the meat quality is also favourably influenced. At one and the same time the development of the muscle fibres must be ensured by optimum feeding.

The basis for the establishment of the first experimental progeny testing stations (fattening and slaughter value examinations) at the beginning of this century, was the fact that the judgement of the conformation of an animal is not a sufficient criterium of its breeding value. Such animals were wished which were able to produce rapidly growing offspring with favourable food conversion and high slaughter value.

It seemed therefore quite natural to investigate the offspring of registered animals belonging to elite herds. At the same time one could naturally make also a selection within the full-sibs of good experimental groups. There is no doubt that so far this selection method has had the greatest significance. Danish investigations have shown (JONSSON, 1966) that selection performed on the basis of full-sib testing is twice or three times as effective as selection based on the progeny testing.

Although progeny- full- and half-sib testing is now more and more replaced by the individual performance testing, I would like to show on some Danish examples that the full- and half-sib as well as the progeny testing, if connected with a considerable concentration of the breeding work and applied in a sufficiently great number of elite herds (inbreeding avoided, sufficiently great genetic variance), are extremely efficient instruments for the improvement of the slaughter value of the pigs.

In the course of the twelve-year period 1957/58—1969/70 the average values of the four Danish official testing stations, where the progeny groups of the state-registered elite breeding farms are tested has changed as follows:

The average thickness of the back fat has been reduced from 3.05 to 2.24 cm (27%).

The average thickness of the side fat has been reduced from 2.75 to 1.70 cm (38%).

The average bacon surface of the loin has been reduced from 38.3 to 23.9 cm^2 (38%).

The bacon surface in % of the total meat surface of the loin has been reduced from 106 to 60 (43%).

From 1966/67 to 1970/71 the meat percentage of the whole body has increased from 59.6 to 61.4.

On the basis of the data of the Danish official fattening performance testing stations JENSEN *et al.* (1970) have calculated the following heritability coefficients:

	h^2
Bacon surface of the back	0.51 ± 0.08
Fat thickness on the back	0.52 ± 0.08
Fat thickness on the side	0.56 ± 0.08
Area of m long. dorsi	0.60 ± 0.08
Meat % + bones in the back	0.61 ± 0.08
Meat % + bones in the ham	0.63 ± 0.08
Meat % in the whole halved carcase	0.60 ± 0.08

The mass of the thickness of the bacon on the back and on the side, as well as of the bacon and meat surfaces do not express exactly the whole meat content of the pigs (PEDERSEN 1968).

If it is important to increase the whole meat content as this is the case in Denmark, then a direct meat content determination of the test animals has to be strived for. As, however, it is impossible, because of economical reasons, to perform a complete parting of the meat, fat and bone of the individual test animals, **Pedersen** (1964) has elaborated the following partial regression method for the calculation of the meat ratios of the test animals on the basis of partial dissection.

$$Y = 18.013 - 0.0765x_1 - 0.07265x_2 - 0.10814x_3 - 0.0121x_4 - 0.0066x_5 + 0.0185x_6 + \\ + 0.0103x_7 + 0.22908x_8 + 0.30556x_9 + 0.26822x_{10}$$

x_1 = slaughter mass, cold, kg
x_2 = back fat thickness over the shoulder, mm
x_3 = fat thickness of the side, mm
x_4 = mass of the leaf dkg
x_5 = mass of the belly dkg
x_6 = mass of the back loin dkg
x_7 = mass of the ham, dkg
x_8 = % of meat + bone in the back loin
x_9 = % of meat + bone in the ham
x_{10} = (for castrated animals = 0, for sows = 1)

The partial dissection gives $R^2 = 0.76$.

It means that 76% of the variance of the total meat mass of the test animals is determined by the partial dissection.

All of the test animals concerned about 22 000 in each year, which are submitted also to feeding test will be judged in two special centres.

In the breeding practice it is most important to concentrate selection to a few, but essential characteristics; since 1st July, 1969, in the Danish pig breeding, these are:

1. Back fat thickness in cm
2. Fat thickness of the side in cm
3. Belly thickness of the flank in cm
4. Area of m. long. dorsi in cm^2
5. Meat ratio of the halved carcase in % (calculated by using the partial dissection)
6. Meat colour

The values for 1, 2, and 3 have to be as low as possible while for 4, 5, and 6 as high as possible.

I am convinced that the full- and half-sib testing combined with the progeny testing will be an effective selection basis, even in the future, for the improvement of the slaughter value of a pig population. However, the individual performance testings give an even greater and more rapid breeding progress than the usual full-sib testing. An absolute condition for the application of the individual performance testing is that the tested characteristic can be measured with equal accuracy when using either of the two breeding methods and it shows one and the same heritability.

When using performance testing characteristics like growth rate and feed consumption can be judged relatively easily, particularly, if the animals are kept under standardized environmental conditions, and fed on a ration containing identical constituents in order to reduce the share of the environmental factors in the total variance. Therefore these tests are often performed in special testing stations to which the animals to be tested are brought by the interested pig breeders. Where there are very big breeding herds including some hundred or even more breeding sows it is naturally possible to set up a special stable for making the performance test on the spot. The same holds of course in respect of the full-sib and progeny testings as well.

The fact that in the course of the past 50 years the full-sib and the progeny testing has become so wide-spread can be explained by the continuously increasing importance of the slaughter quality in pig production. Earlier when using the full-sib and progeny testing methods the slaughter quality could be determined only after slaughtering the test animals and then the results could be used in the selection of the breeding animals.

However the individual performance testing has achieved more and more importance during the past few years as it offers better possibilities for the judgement of the slaughter quality in living animals.

By using ultrasonic technique the thickness of fat layers can be measured with sufficient accuracy not only on the back line but on the sides too. However the measurement of the back fat thickness as already mentioned, is not a reliable expression of the total meat content of the pig. Still we may hope, that the future development of the ultrasonic technique will make it possible very soon to measure with sufficient accuracy not only the total bacon thickness of the back but at the same time the mass of the back musculature (m. long. dorsi and m. multifidus dorsi) as well. The surface of the m. long. dorsi, however, is not in a close correlation with total meat content. PEDERSEN (1964) found a phenotypic relationship between the surface of the m. long. dorsi and the meat ratio of the tested pigs $r = +0.5$.

In a bacon producing country like Denmark an increase of the surface of the large back muscle is of great importance.

By the use of a new ultrasonic equipment we received repetition coefficients of 0.91 and 0.97 when measuring the muscle surface area and the bacon thickness on the side of the living pigs, respectively. The correlations between the measurements made in living animals and on the photographs taken of the same pigs after slaughter were as much as 0.69 and 0.82 for the muscle surface area and the bacon thickness of the side respectively. Before measuring the pig is anaesthetized.

It is not satisfactory to make the selection of the breeding animals on the basis of the meat content that is calculated exclusively from the results of the individual performance test.

The meat quality of the individual animals must also be determined.

At present a most extensive research focused on the meat quality is being done in many countries. Unfortunately we are lacking both a reliable definition for the meat quality and the necessary instructions with what the quality can be determined.

In Denmark STAUN—CHRISTENSEN (1968) have proved that by means of a small operation it is not too difficult to get a sample from the great back muscle of a narcotized pig. The samples' weight is about 25 g and this amount of meat is enough for the determination of the diameters of muscle fibres, their number per mm^2, the length of the muscle fibres, the water binding capacity, the pH value, the protein content, the dry matter and the intramuscular fat respectively.

As up to the present neither the meat ratio nor the meat quality are measurable with sufficient accuracy in live animals, the individual performance testing must be probably combined with full-sib testing in the following years.

In various countries, at any rate in Denmark, there was a fear, that a spreading of infectious diseases might occur if the boars are taken back to the pig breeding farms from the individual performance testing stations. By using artificial insemination, this risk can be reduced considerably. At the same time artificial insemination increases the utilization of the very great selection pressure applied at the performance test.

Non-castrated boars

The castration of the boar causes the reduction of the meat production and an increase of fat production. This has been proved in many experiments. The data of Table 4 are the results of a Danish experiment, where in each repetition 3 full-sibs were available i.e. a male, a castrated male and a female.

It was of very great importance for the meat content of the products to be sold if the castration of the boars could be avoided. This is, however impossible, because of the sex odour (boar odour) occurring in the case of boars. In boars slaughtered at similar live weight an extremely great variation in the boar odour, which is independent of the age of the boar, has been established (Jonsson 1967). It is therefore an important question whether the intensity of the boar odour is genetically determined, if this is the case then it can be eliminated by selection. On the basis of a material, including 1907 boars from Danish boar stations a heritability coefficient for the boar odour of 0.14 ± 0.08 has been established by Jonsson *et al.* (1969).

Table 4

Effect of the castration on the meat and fat production

	Boars	Castrated boars	Sows
Number of animals	16	16	16
Daily weight gain g	743	692	721
Feed unit per kg weight gain	2.80	3.09	2.89
Back fat thickness, cm	2.66	3.17	2.75
% meat of the pigs (determined by full parting)	61.3	56.2	60.0

Conditions: growing period from 20—90 kg live weight;
individual feeding;
very good housing conditions.

As already mentioned perfect utilization of the genetic basis for the meat production of pigs depends on whether the feed contains sufficient amount of proteins with the necessary levels of certain essential amino acids. Animal foodstuffs like blood meal, fish meal, wale meal, casein, skimmed milk powder and meal have a relatively high content (7.25—9.269 pro 16 g N). In meat bone meal and buttermilk powder these values are about 5.9 and 5.7 respectively. In feeds of plant origin the lysin content is considerably lower and fluctuates from 1.83 in milocorn to about 6.0 in soy bean grits.

One has to calculate with the fact that the ratio of foodstuffs of animal origin will be less than at present during the following years, partly because a certain part of it (fish- and milk protein) will be increasingly used for humane nourishment and partly because no considerable increase of the amount of animal foodstuffs can be expected. In certain pig producing countries there are no or only very restricted possibilities for using foodstuffs of animal origin in pig feeding.

As already mentioned lysin is a most important factor in the utilization of a good genetic background of animals as regards meat production. For ceasing a possible lack of lysin, there are as far as I can judge it, only two possibilities: either the rations have to be supplemented with industrially produced amino acids (microbial or synthetic) or vegetable foodstuffs with a higher lysin content must be found.

The economical effects of the first possibility are considerable as the price of the technically produced lysin (as well as that of the methionin) is so low at present, that the lysin, under certain conditions i.e. high prices of milk and meat products, soy bean grits as well as of pigs with intensive meat production, must be taken into consideration when calculating the best and less expensive composition of pig rations.

How intensively the need of industrially produced amino acids will increase during the coming years is depending on the development of the second possibility i.e. what an amount of vegetable foodstuffs with high amino acid contents will be found.

Until before several years, all over the world the plant breeders were striving for an increase of the average yield per surface unit without paying attention to the composition of the cropped products (except the water content).

In the past years, however, there have been found new varieties of maize, wheat and barley containing considerably more protein with higher levels of amino acids, that are important for the growth and meat production.

If this development will be continued, then it can be expected that the imminent problem of producing enough protein for the increasing mankind and the animal production will be somewhat reduced at any rate in the course of the coming decades.

But there is also one more possibility, although its importance seems to be smaller to search for already known varieties of plants with relatively high lysin content and to take them into cultivation. These are plants which earlier, owing to their requirements for special conditions, have been excluded from cultivation.

In Denmark, during the past years, we have repeatedly started cultivating the *Vicia faba*, as our investigations had shown that these beans had a relatively high lysin content (about 6.25 g per 16 g N). In comparison to this soy bean grits contains about 6.0 g lysin per 16 g N. Extensive feeding experiments have proved that the *Vicia faba* can be added to the ready-made feed mixtures in an amount of 10—20%. The up-to-date protection against parasites (plant-louses) has made possible again the cultivation of *Vicia faba* (Hansen *et al.* 1969.)

However, *Vicia faba* cannot be fed to breeding sows as the littersize will be reduced and the dry period of the sows will be prolonged.

We are conducting now a series of feeding experiments with peas that have also rather high lysin contents (6.93 g lysin per 16 g N; *Vicia faba* 6.25; soy bean grits 6.0) as it seems likely that a number of varieties of peas, having less crop-loss when harvested by combines, will be marketed. Also in this field, we are facing a valuable development.

The further development of the production and marketing of the "technical meat" produced from soy bean protein is an imminent cloud over the future development of the pig production.

The development cannot be stopped, and its extension cannot be predicted.

In my opinion, however, this artificial meat will replace the fat and low-quality meats, while fatless first-quality pork can be sold also in the future. This is one more basis for my belief that "the pig of the future" will not be by all means the one which is produced at the lowest rate, but rather the one with high meat ratio and good meat quality.

Millions of pigs with very bad carcase value are slaughtered in the world every year, as the breeding plans have not been based on a selection concentrated on animals with a genetic basis for the production of much and good meat.

But also millions of pigs with a good genetic basis for meat production are slaughtered in the world every year, which could have produced also a good slaughter value, which however never received the chance for this, as they were fed unadequately. In none of the cases was this the pigs' fault.

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FORUM

DAILY AND ANNUAL RHYTHM OF WATER REGIME INDICES IN PEACH VARIETIES OF VARIOUS RIPENING TIME

The following observations have been made concerning water regime in peach varieties of various ripening time. Transpiration is more intensive in younger than in older leaves. At the beginning of the vegetation period, with an identical water supply, transpiration is the most intensive in the variety Julia. In the daily rhythm of transpiration Julia is the first to attain a maximum; towards noon transpiration decreases in all the three varieties due to the absence of turgor. The water saturation deficit measured at different parts of the day is in inverse correlation with the water potential. At noon, with total insolation, the temperature of the leaves suffering from a water saturation deficit is 4—5°C higher than at the stage of total turgor. Owing to a temporary water deficiency occurring in the leaves as a consequence of increased transpiration the amount of free water decreases and that of combined water tncreases. There is a negative correlation between the stoma density and stoma dimensions if the leaves in the varieties examined. The nearer the moisture content of the soil approaches ois total water capacity the higher is the number of open stomata.

Introduction

Each life process of a plant can only take place properly if the water supply in the cells and tissues is satisfactory. Under Hungarian weather conditions the overwhelming majority of plants, including nearly all cultivated fruit species, is often exposed to the danger of water deficiency. Even the peach — generally though incorrectly believed xerophilous — yields 20—40 per cent more when irrigated than when grown without irrigation (Mohácsy Sen.—Maliga—Mohácsy Jun. 1967).The development of plants is in fact controlled by the internal water balance and turgor prevailing in the cells. It is these factors that mainly determine the physiological processes and conditions that influence the extent and quality of development and eventually result in high yields.

Our experiments were aimed at studying the water regime in peach varieties of different ripening time during the vegetation period. As a further objective we shall study the course of shoot growth and fruit development in these varieties as reflected by the most important indices of water regime, under irrigated and unirrigated conditions.

Material and Method

In two successive years (1968—1969) the daily and annual rhythm of the water regime indices was studied in three peach varieties (Győztes, Júlia, Elberta) of different ripening time.

The experiment was carried out at the Cserszegtomaj experimental site of the Keszthely Agricultural University, in the field with six replications. The examinations were made on eight year old trees grafted to bitter almond stocks. Care was taken to perform the examinations of the water regime indices with healthy leaves of identical age and exposure, taken from the same leaf storey, at the same time, under similar conditions.

The transpiration intensity was measured with a thermistor transpirometer according to the method of BORKA (1967) and PARCEVAUX (1964). The forms of water in the leaves were determined by the method of GOLODRIGA (1965) and GUSEV (1965), the absorption value with that of ÖNÁL (1964), the saturation deficit with BORKA's (1969) and the course of waterloss in the plant with PARCEVAUX's (1964) method. The density and dimensions of the stomata were studied with a microscopic, while their openness with a collodial method.

Results

In the various varieties the trend of water regime indices measured in different periods was determined partly by climatic factors, partly by the developmental stage characteristic of the variety. E.g. in the variety of which the leaves were at the stage of elongation and growth the total water content of the leaves, and the proportions of water forms within them, were different from those in a senescing leaf, due to a more intensive metabolism and the dominance of hydrophilous colloids in the protoplasm. (Transpiration is related to the total water content of plants; waters fixed the least are the first to evaporate, and colloidally or osmotically fixed forms of water are moved only in the absence of free water.) Consequently, the indices of water regime — transpiration intensity, saturation deficit water fractions — must be studied in leaves of the same age. As it is known, evaporation shows a close correlation with the age of the organs (CATSKY 1965, PROKOFYEV 1963, GEJ 1962, KUSNIRENKO 1967). The unit area of the first leaf developed at the beginning of the vegetation period for example evaporated 46 per cent of the free water evaporation under the same climatic conditions, while leaves taken from the tenth storey evaporated 81 per cent (Table 1).

The young leaves were less xerophilous than the older ones. According to ASHBY—WANGERMANN (1950) the shape and size of cells in successive leaves show a definite gradient. (The decrease in the turgor stops and unfavourably modifies the growth and development of cells.)

At the beginning of the vegetation period, with a sufficient soil water content the variety Julia showed the highest transpiration. The water loss in the varieties Győztes and Elberta was about the same (Table 2).

On the average of several measurements the daily rhythm of transpiration showed a similar trend. The maximum was first attained by the variety Julia; between the varieties Győztes and Elberta no significant difference was found (Fig. 1).

In peach — as in other cultivated plants (BORKA 1969) — a certain decrease was observed at noon in the daily course of transpiration, caused by the reduced turgor (PORPÁCZY 1964). According to some authors the decreased assimilation is the very consequence

Table 1

Percentage evaporation of peach leaves of different insertion

Evaporation of free water	Transpiration of first leaf	Leaf surface tenth leaf
100	46	81

Table 2

Percentage transpiration of the peach varieties Győztes, Júlia and Elberta at the beginning of the vegetation period, with a soil water capacity of 70 per cent

Evaporation of free water	Transpiration of leaf surface		
	Győztes	Júlia	Elberta
100	65.3	73.3	66.0

of the saturation deficit (LORENZEN 1957). In spite of the abundant water content of the soil the water balance was disturbed owing to the intensive insolation prevailing at noon, high temperature and low relative moisture content. The plant was not able to take up as much water from the soil as it evaporated into the air through its leaves. The result was a stagnation in the course of transpiration at noon, which was only compensated around 3 p.m. when insolation decreased and turgor improved. In the morning hours the leaves were in full turgor, transpiration was determined exclusively by climatic factors (air and soil temperature, air movement, relative moisture content of air). Later on, around noon, when the turgor decreased the water deficiency in the leaves increased and to some extent inhibited a "stressed" water loss induced by the climatic factors. Under the influence of the mentioned factors there occurred a water deficit even when the soil contained abundant water. Water deficiency in the plant not only depends on the water supply from the soil but also on the extent of water loss. A temporary water deficiency occurs even when sufficient soil moisture is available, if evaporation from the plant is too rapid. On the other hand, a relatively dry soil may provide sufficient water if the water consumption of the plant is low (POSPISILOVA 1966, KRAMER 1963).

The water potential as well as the water saturation deficit in the leaves were examined at different periods of the day. Similarly to the results of other authors (POSPISILOVA 1969, SHINN—LEMON 1968) the water saturation deficit was found to be inversely proportional with the water potential (Table 3). According to Weatherley's model investigations this phenomenon is expressedly caused by the low water conductivity of the soil (WEATHERLEY 1963).

According to the data measured with a thermistoric point thermometer leaf temperature was 5°C higher when the leaves were exposed to direct insolation and suffered from a water saturation deficit (in the absence of transpiration) than when in full turgor (Table 4).

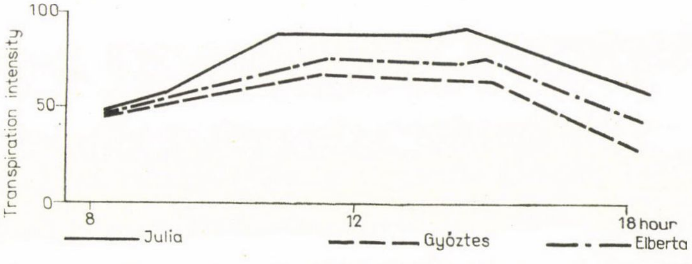

Fig. 1. Daily rhythm of percentage transpiration intensity in different peach varieties at the early phase of vegetation, with a soil water capacity of 70 per cent

Until the leaves contained sufficient quantities of "mobile" water to transpire intensively, the temperature difference between the leaves exposed to total insolation and those shaded was maximum one or two degrees.

Petinov—Razmaev (1961) did not find any change in the total water content with increasing leaf temperature in wheat and maize, however the amount of colloidally fixed water increased. In agreement with observations made by some authors (**Tanner** 1963, **Stalfelt** 1962) we found that in the case of a sufficient water supply an increased temperature always resulted in more intensive transpiration.

Table 3

Daily rhythm of water saturation deficit in the peach variety Győztes

Time	mg weight of 8 m^2 leaf	
	fresh leaf	after 60 minutes (with 100% moisture content)
at 8 a.m.	158	160
at noon	151	159
at 6 p.m.	154	160

Table 4

Temperature of leaves saturated with water and suffering from water deficiency respectively, in the peach variety Júlia

In sunshine	In shade
24°C	19°C

Table 5

Percentage proportions of water forms in the peach leaves at the beginning of the vegetation period, with a 70 per cent soil water capacity

Water fractions	Győztes			Júlia			Elberta		
	hours								
	8 a.m.	12	6 p.m.	8 a.m.	12	6 p.m.	8 a.m.	12	6 p.m.
Free water %	16	11	14	19	14	15	14	9	11
Fixed water %	84	89	86	81	86	85	86	91	89

After a certain maximum attained, the turgor ceased in the leaves and the total water content decreased. According to **Tew—Taylor—Aschroft** (1963) transpiration increasing in a direct ratio with the temperature is not influenced even by a relative moisture content ranging between 35 and 65 per cent, but only by the temperature.

As a consequence of water deficiency the proportions of water forms in the cells and tissues changed. While in the morning all three varieties contained considerable quantities of "free" water, at noon — as a result of the loss of "free" or "mobile" water — the proportion of fixed water increased, and the original status was only re-established towards the evening, when water supply improved (Table 5).

With sufficient soil moisture available, the effect of insolation on transpiration was not the same in the three peach varieties. When the sky was overcast for at least 60 minutes, the decrease in transpiration was more intensive in the variety Győztes than in Julia or Elberta (no difference was found between the two latter varieties) (Fig. 2).

At the same time, the water saturation deficit in leaves not directly exposed to insolation was found to be lower than in those exposed to direct sunshine. Shinn—Lemon (1968) observed the same with agricultural cultivated plants.

We examined the stoma density and stoma dimensions in the above three varieties and in a seedling of the same age. Each tree had characteristically hypostomatic leaves (Fig. 3). Of the three varieties Győztes had the highest number of stomata per unit area, while the stomata were the smallest in size (Table 6, Fig. 3).

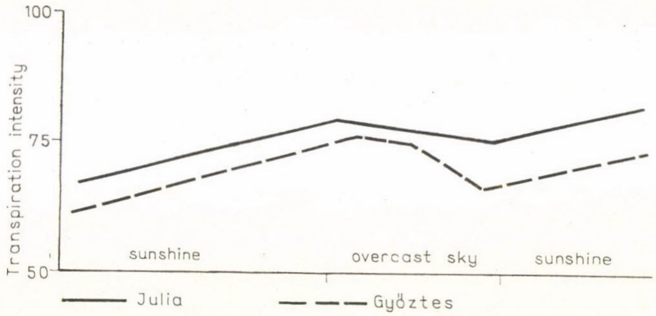

Fig. 2. Effect of insolation on transpiration in the leaves of different peach varieties, wit a soil water capacity of 70 per cent

According to some authors (Wit 1958, Parcevaux 1964) the highest resistance in water movement occurs when the water molecules enter the atmosphere through the stomata. The smaller the stomata the higher the possibility of vaporization.

Stoma density was the lowest while stoma size the largest in the variety Julia. The highest number of stomata per 1 cm^2 was found in the variety Győztes but their size was the smallest. Elberta was between the two former varieties regarding its number and size of stomata. Thus, in the three varieties examined a negative correlation was found between the number and size of stomata. At the same time the leaves of a seedling were also examined: it was here that the number and size of stomata were found to be the smallest of all (Table 6, Fig. 3). In order to draw any conclusion from what has been said above new and more extensive investigations are required.

The openness of the stomata on the leaves was examined with a collodium coating. The nearer soil moisture approached the total water capacity, the higher was the number of

Table 6

Number of stomata per unit area and stoma size in peach varieties

	Győztes	Júlia	Elberta	Seedling
Stoma number/m^2	19545	11734	12936	9894
Stoma length (micron)	29.90	43.24	33.58	24.10

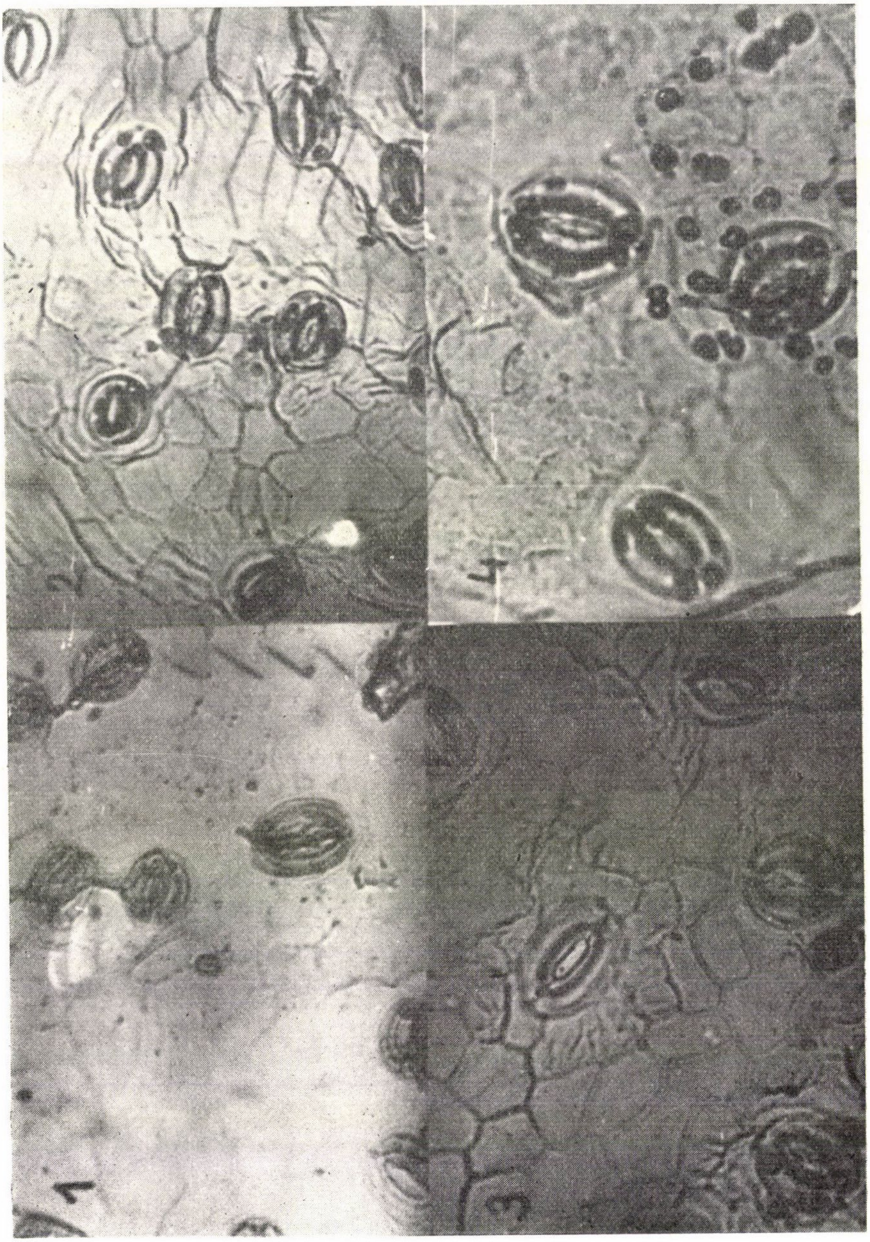

Fig. 3. Number of stomata per unit leaf area and stoma size in peach varieties

Table 7

Percentage intensity of transpiration in the three phases of fruit development

Evaporation of open water surface	Transpiration of leaf surface								
	Győztes			Júlia			Elberta		
	1	2	3	1	2	3	1	2	3
100	65.3	93.4	51.2	73.3	96.7	68.1	66.0	75.1	94.2

open stomata found on the leaves. The low relative humidity, the water saturation deficit occurring around noon resulted in most of the stomata being closed through a hydroactive reaction. It must be mentioned that the leaves at the base of the shoot were closed earlier than those on the top, probably because younger leaves detracted the water from the older ones due to their more intensive metabolic processes.

In the development of peach three phases are distinguished (Mohácsy Sen.—Maliga—Mohácsy Jun. 1967). The first one is the phase of stone formation. In this phase the only difference between the water regimes of the three varieties was the higher water utilization — as a genetic character — of the variety Julia. As we have seen before, in the initial phase of the vegetation period, with sufficient soil moisture available and under identical edaphic and climatic conditions Julia required and transpired the largest quantity of water.

The second phase of development is characterized by the hardening of the stone. According to measurements made it was in this phase that the highest amount of water was used by all varieties examined. E.g. the transpiration intensity of the variety Győztes was in the first phase about the same as that of the Julia, but several days later — when stone hardening began — it was 20 per cent higher (Table 7).

The increased water utilization by the variety Győztes only lasted for about 5—6 days then decreased again (Fig. 4).

In the third phase of development the transpiration did not increase. According to literary data the water uptake by the trees in this phase is also high, due to the intensive transpiration of the fruits. For methodological reasons we could not measure the transpiration of the fruits, and only concluded on the water utilization of the varieties examined from the transpiration of the leaves.

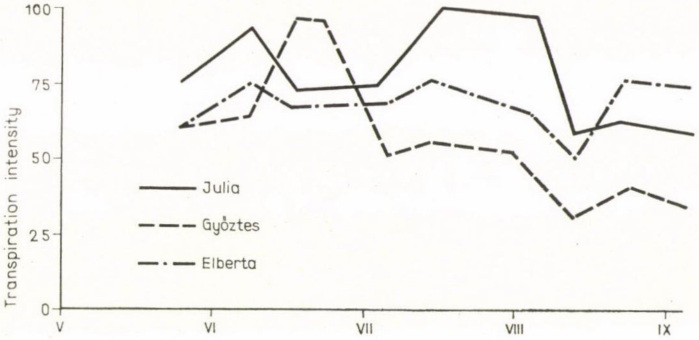

Fig. 4. Annual rhythm of transpiration in peach varieties with different ripening time under identical climatic and edaphic conditions

Table 8

Percentage water content and forms of water in the leaves of the peach varieties Győztes and Elberta

Water fractions	Győztes	Elberta
Total water content	67	80
Free water	6	13
Fixed water	94	87

After the phase of stone hardening the transpiration in the leaves of the variety Győztes was only half of that measured earlier. Julia was the following variety to enter the phase of stone hardening. It is worth mentioning that in this period, when the dry weather lasted for weeks, transpiration was very low in all three varieties in spite of the high insolation and temperature and low relative humidity. Under such dry conditions the variety Julia was not able to produce the increased water uptake corresponding to its phase of development either. However, 24 hours after a precipitation of 6 mm, when the upper layer of the soil attained 88 per cent of the total water capacity, the transpiration of Julia considerably increased. At the same time, transpiration in the variety Győztes did not increase significantly in spite of a satisfactory water supply, because the plant had already got through the phase of intensive water utilization.

From the middle of July till the beginning of August the weather was dry again, when turgor was not re-established in the leaves even during the night. As a consequence no increased water utilization characteristic of the phase of stone hardening could be observed in the latest variety: Elberta. The vegetation period of the early variety was found in both years to end much earlier than that of a later variety. E.g. at the beginning of September the variety Győztes showed but a minimum water requirement (its transpiration was 57 per cent of the evaporation). At the same time the water utilization of the late variety Elberta was found to be still very intensive: transpiration was 89 per cent of the evaporation (Fig. 4).

The data of water potential measurements made in the foliage leaves unambiguously confirmed the above. The total water content determined in the leaves of the variety Győztes at the beginning of September, and the ratio of water forms within showed — as opposed to the variety Elberta — a considerable slowing down in the metabolism (Table 8).

The physiological status of the above two varieties is well reflected by the absorbing ability of the cells and tissues — shown as plotted against time in Fig. 5.

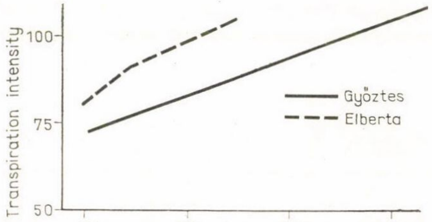

Fig. 5. Percentage water absorbing ability of the peach varieties Győztes and Elberta in the function of time

The investigations disclosed that in the course of their development the total water content of plants decreases. At the same time, with the reduced process of synthesis, changes related to the hydratation of colloids take place in the plasm and consequently the hydratation degree of the protoplasm colloids gradually decreases resulting in a slowing down of the active physiological processes and an increase in the decomposing enzymes.

The water uptake by the plant is a process highly dependent on the metabolism which takes place through direct and indirect relations at the expense of respiration energy. Senescence upsets the balance between synthesis and decomposing processes, energy reserves become more and more depleted, and the plant eventually arrives at the period of dormancy.

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CONTRIBUTIONS TO THE PAPER OF ZATYKÓ, J. M.: "EFFECT OF BENZYLADENINE ON THE AMOUNT OF LEAF PIGMENTS IN BEAN" PUBLISHED IN THIS PERIODICAL, *20* (3–4) 425–437.

WHY IS IT ONLY IN THE DARK THAT BENZYLADENINE IS EFFICIENT IN MAINTAINING THE CHLOROPHYLL CONTENT?

The subject is by no means new, but new facts can be told about it all the same, e.g. that the "feedback" system ensures the chlorophyll–carotenoid balance. This very interesting statement made by Zatykó with due cautiousness is — unfortunately — not convincing. In my opinion, changes in the balance of the pigment systems in leaves kept in light and darkness, respectively, can be better explained by the changing stability of protein in the pigment + protein complex. The polar chlorophyll molecule in the thylakoids is bound to structure proteins. The carotenoid molecules are also located in the thylakoids, though in a different bond. If the skeleton of this system: the protein is attacked by catabolic processes becoming dominant in the dark, or without the regulating effect of the root, the complex of the different pigments and the proportion of the compartmentalized parts will, naturally, be disturbed.

Zatykó's data are worth paying attention because they give information about unexpected facts: it was only in the dark that benzyladenine was efficient in maintaining the chlorophyll content! Does this observation — by any chance — affect some photochemical interaction?

Of course, the paper was sent to me as a manuscript and the ultimate form of the tables had not completely developed then. For this reason I had some difficulties in getting acquainted with the question. I should have been glad to see the dynamics of the phenomena in graphic representation. I admit too, that I do not quite see whether figures written with ± signs after the mean values are values of standard deviation or other indices of mathematical statistics. They generally appear to be extremely high.

Furthermore, I do not understand the figures concerning the total xanthophylls. Namely, the totalled values of neoxanthin, violet-xanthine and luteine are not in this column, although they are in fact xanthophylls as they are oxidized compounds! All this is — of course — a question of convention which, however, must by all means be taken in consideration.

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IS ABSCISIC ACID FORMED AS A RESULT OF THE LIGHT MEDIATED DECOMPOSITION OF CAROTENOIDS?

I have read with interest the contribution "Effect of benzyladenine on the amount of leaf pigments in bean" published by J. M. Zatykó in Acta Agronomica.

The problem of the action of cytokinins in retarding senescence and degradation of pigments is not a new one but the present paper is original by the fact that it considers carotenoids jointly with the chlorophylls and both in the dark and in the light. Studies on carotenoids regarding this problem are relatively poor in the literature.

The paper and the results are clearly presented and the discussion raises many important questions.

I regret, however, that the "feedback" system mentioned by the author to explain the data was not developed a little more.

Another point, regarding carotenoids and exposure to light has not been discussed in the paper; it is the formation of abscisic acid and related growth inhibitors as a result of the light mediated decomposition of carotenoids (TAYLOR and coll., 1967, 1968, 1970).

This promises new development in the comprehension of senescence through hormonal interactions.

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DOES BENZYLADENINE HAVE ANY EFFECT ON THE DECOMPOSITION OF CHLOROPHYLLS IN ILLUMINATED BEAN LEAVES?

The paper by J. M. Zatykó on "Effect of benzyladenine on the amount of leaf pigments in bean" deals with a fairly neglected area of plant hormone research. As also emphasized by the author, it is hard to understand why earlier workers have paid so much attention to the study of chlorophyll level as affected by cytokinins and did not study simultaneously the effect of these plant hormones on the carotenoids, the physiological function of which is gaining more and more attention in other areas of plant physiology. The very carefully conducted experiments of Zatykó clearly and convincingly demonstrate that benzyladenine really affects the level of carotenoids and also provides evidence that this effect is different from that exerted on chlorophylls. It is also significant that not all carotenoids are affected by benzyladenine to the same extent. Another important observation is that the effect is dependent on the light conditions. It is somewhat surprising, however, that the author did not observe any effect of benzyladenine on the decomposition of chlorophylls in illuminated bean leaves. Light itself, indeed, has in many systems a "cytokinin-like" effect, on the other hand, however, light is often needed for a full effect of cytokinins. Therefore, this part of the work appears to need confirmation, perhaps by using different light and cytokinin levels.

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CAN BENZYLADENINE POSSIBLY CONTROL THE LEVEL OF ENDOGENOUS INHIBITORS?

Details concerning the manuscript: 1. Do seedling cuttings have juvenile leaves or trifoliate ones? What was, precisely, the object of analysis? 2. Did benzyladenine have any effect on the growth of the cuttings? 3. It should be noted what the data are followed by; Standard deviation or standard error?

General comments

Tests carried out by Dr. Zatykó revealed that benzyladenine (BA), one of the synthetic compounds showing a cytokinin-like activity (LETHAM 1967), when applied to *Phaseolus vulgaris* L. cv. Fertődi seedling cuttings comprising the juvenile leaves (1) retards chlorophyll degradation in the cuttings held in darkness, and is ineffective in the case of explants kept in permanent illumination. Irrespective of light conditions BA (2) did not prevent the drop of total carotenoid content in the leaves. However, the cytokinin (3) accelerated neoxanthin disappearance in the leaves held in darkness, being in this respect ineffective when the cuttings were maintained in light.

Each of these facts is worth separate discussion. The first finding is not unexpected. It confirms several previous reports that cytokinins effectively arrest yellowing of the leaves of different plant species in darkness. This fact was for the first time established for *Xanthium pennsylvanicum* (RICHMOND—LANG 1957, OSBORNE 1962) and then confirmed to be true for tobacco (MOTHES *et al.* 1959, SUGIURA *et al.* 1962; cf. reviews by MOTHES 1964, OSBORNE 1965), bean (LEOPOLD 1964), *Rumex* (GOLDTHWAITE—LAETSCH 1968), nasturtium (BEEVERS 1966), *Brassica* (ABRAMS—PRATT 1966), oat (GUNNING—BARKLEY 1963), rice (YAMADA *et al.* 1964), barley (KENDE 1964), maize (LEOPOLD 1964, TAVARES *et al.* 1968), *Taraxacum* (FLETCHER—OSBORNE 1965), and others. Most of the quoted tests were carried out with excised leaves, de-rooted explants or leaf discs kept in darkness or in dim light. In such conditions chlorophyll synthesis is hindered the leaves turn rapidly yellow and the retarding effect of cytokinins on leaf senescence is visualized. The response of some plant species is very selective and sensitive; on this basis several biotests for cytokinins have been elaborated, using leaf discs of *Xanthium* (OSBORNE—MCCALLA 1961), *Raphanus* (KARANOV —NEICHEVA 1970), or sections of the first barley leaf (KENDE 1964). The latter biotest can also be conducted in light; but in this case the test is more prolonged in time (cf. KULAEVA 1966).

In contrast to tests carried out with detached leaves, leaf discs or de-rooted explants, usually no visual effect of cytokinins on senescence (measured as the progress of yellowing) was found with leaves attached to intact plants (cf. KULAEVA 1962, ENGELBRECHT 1964), since such leaves did not yellow during the test period. The situation is quite different with the juvenile, non-trifoliate leaves of bean. In primary bean leaves the gradual loss of chlorophyll begins soon after the expansion growth of the leaf blade has stopped. Yellowing for a few days before abscission is the most conspicuous change accompanying senescence of these leaves (cf. LEOPOLD 1964). Treatment of the primary bean leaves with BA retards their senescence both on the intact plant (FLETCHER 1969, JACOBY—DAGAN 1970) and on the de-rooted explants (LEOPOLD 1964) kept in 14—16 h photoperiod.

In the case of intact Red kidney bean plants visible symptoms of senescence of the primary leaves in the control appeared approximaly 5 weeks after sowing (FLETCHER *et al.* 1970). Detailed studies carried out by JACOBY—DAGAN 1970) revealed that primary leaves of bean cv. Brittle Wax, grown at 16 h photoperiod at 22—25°C, became yellow and abscissed at the age of about 28 days, whereas the BA treated ones were green even at the age of 50 days. Growth rate of the leaves was initially linear until the age of 11—12 days, then decreased and stopped at the age of 17—18 days. BA did not affect the initial growth rate, but extended the leaf growth period until the age of about 20 days after sowing. As a result of this the BA-treated leaves had a markedly increased area in comparison with the control, untreated ones. In the control leaves the chlorophyll content gradually increased as long as the leaves expanded. Soon after the cessation of the leaf expansion, i.e. at the age of about 17 days, the chlorophyll loss started. Benzyladenine reduced the net chlorophyll synthesis in the case of young (9-day-old) seedlings, but afterwards the same rate of chlorophyll synthesis as in the vigorously expanding control was regained. At the age of 16 days the net chlorophyll accumulation was stopped; at this age the BA treated leaves have a somewhat higher level of chlorophyll compared with the untreated control. In the latter ones, however, the loss of chlorophyll started, whereas BA prevented this process at least until the age of 27 days. Effects of BA on growth and chlorophyll content of primary leaves were similar to those induced by decapitation of the seedlings above the primary leaves (cf. JACOBY—DAGAN, 1969, PHILLIPS *et al.* 1969).

Seedling cuttings of bean cv. Fertődi, maintained at permanent illumination, showed visible symptoms of senescence and lost about 50 per cent of the original chlorophyll content (as measured at the age of 8 days, that is at the beginning of the treatment period) at the age of about 18—23 days (cf. Material and Methods in Dr. Zatykó's paper). Hence, they behaved in a similar manner as the intact plants of bean cv. Brittle Wax (cf. JACOBY—DAGAN 1970) and as the de-rooted bean cuttings in Leopold's tests (LEOPOLD 1964, LEOPOLD—KAWASE 1964). However, the results presented in Tables 3 and 4 of Dr. Zatykó's paper are in contrast to those reported by LEOPOLD (1964), FLETCHER (1969), JACOBY—DAGAN (1970) and others. Roots are suspected to be organs synthesizing cytokinins (cf. KENDE 1965) and they have a marked retarding effect on leaf senescence (cf. KULAEVA 1962, KENDE—SITTON 1967, SITTON *et al.* 1967, WAREING—SETH 1967). Since Dr. Zatykó used de-rooted cuttings, the retarding effect of BA on primary leaf senescence should clearly be demonstrated. The negative result might have been caused either by the different response of the cultivar Fertődi to light and BA in comparison with the response of other bean cultivars, or by a too low concentration of applied BA. The latter explanation seems to be quite probable: Dr. Zatykó used BA at the concentration of 5 mg/l, applying it by spreading over the leaf, whereas in all the other quoted studies (LEOPOLD 1964, FLETCHER 1969, JACOBY—DAGAN 1970) BA was used at the concentration of 30 mg/l and was applied by painting the leaf blade. Since Dr. Zatykó's tests also differed from the quoted tests by other experimental details (e.g. permanent illumination), the reason for the ineffectivity of BA in arresting the chlorophyll disappearance in light cannot be found without the performance of additional tests.

Although BA retarded the chlorophyll loss from the bean leaves kept in darkness (Table 1) it did not affect the rate of carotenoid disappearance. This is in constrast with the results of the tests carried out with *Salvia* (GASPAR—XHAUFFLAIRE 1968) and *Nicotiana* (AVUNDZHYAN—ARUTYUNYAN 1969) leaft discs, in which BA maintained a high level of both carotenoids and chlorophylls, the preserving effect on the carotenoid level being markedly more pronounced than that on the chlorophyll level. Another cytokinin, kinetin, also retarded chlorophyll and carotenoid degradation in *Raphanus* leaf discs (KARANOV—NEICHEVA 1970). Generally, the carotenoid content is correlated with the chlorophyll content, especially in young and expanding leaves (cf. VALADON—MUMMERY 1969). However, since the pigment synthesis is controlled both by genetic (albino mutants) and environmental factors (cf. HIPKE 1971, MAHLBERG—VENKETESWARAN 1966), this correlation is not always strict. The correlation is lost in senescing leaves in which chlorophyll disappears without a concomitant disappearance of the yellow pigments. The ratio of chlorophylls to carotenoids can also be altered by some chemicals like gibberellin (SZALAI 1968, 1969), growth retardants (KNYPL 1970a) in the sense, that chlorophyll synthesis in blocked without concomitant inhibition of carotenoid synthesis. This means that the correlation only exists in one direction: The tissue containing much chlorophyll as a rule also contains much carotenoids, but the tissue with a high level of carotenoids may be free of the green pigments. Two groups of hypotheses have been evolved to explain the constant ratio of chlorophylls: carotenoids in green tissues: (1) carotenoids may be the precursors of the chlorophyll molecule, especially of a phytol side chain (SAAKOV 1968, LOSEV—SHLYK 1969), and (2) carotenoids can serve as protective agents against lethal photosensitized oxidation of the chlorophylls (cf. a review by KRINSKY 1966, and references quoted by Dr. Zatykó). According to the latter hypothesis the epoxide cycle plays an especially important role. Chlorophyll molecules, excited by light, are normally de-activated in the process of photosynthesis. Occasionally, however, the excited chlorophyll molecule combines with molecular oxygen which can lead to its oxidation. This potentially lethal combination is inactivated by a carotenoid such as zeaxanthin which itself oxidizes to its epoxide, antheraxanthin. The latter compound, on the other hand, is regenerated to the protective substrate, zeaxanthin, by an enzymatic reaction catalyzed by antheraxanthin, de-epoxide (cf. Fig. 4 in Krinsky's paper). The epoxide cycle has been postulated to operate in photosynthetic bacteria and in green algae; there is also some evidence that it operates in higher plants (cf. SAAKOV—NASAROVA 1970) but this has not yet been conclusively proved.

Dr. Zatykó's paper does not provide new conclusive evidence bearing reports that destruction of carotenoids takes place in bean leaves held in darkness, especially of carotenes, lutein, violaxanthin and neoxanthin (BANDURSKI 1949, VALADON—MUMMERY 1969). However, the fact that benzyladenine accelerated the loss of neoxanthin in darkness is highly interesting, although this cannot be satisfactorily explained at present. The physiological role played by the yellow pigments is far from being understood. ARONOFF—KIRK (1967) postulated that xanthophylls, especially derivatives of neoxanthin and violaxanthin, can regulate the extent of aggregation of the chlorophylls in chloroplasts. On the other hand neoxanthin, violaxanthin and related epoxides produce growth inhibitors on illumination (TAYLOR—BURDEN 1970a; b, BURDEN—TAYLOR 1970); one of these inhibitors, extracted from bean and wheat seedlings was as effective physiologically as abscisic acid which has been shown to be a potent inhibitor of chlorophyll synthesis (cf. LICHTENTHALER—BECKER 1970). The content of neoxanthin and lutein epoxide rises in plants subjected to low temperature stress (SAAKOW 1970). It is thus quite possible the fact, revealed by Dr. Zatykó, that benzyladenine accelerates destruction of neoxanthin, has great physiological significance. It has been postulated that neoxanthin can be synthesized from antheroxanthin in darkness (cf. VALADON—MUMMERY 1969) as an alternative derivative to violaxanthin. Benzyladenine, by accelerating the destruction of neoxanthin may control the level of endogenous growth inhibitors like abscisic acid (cf. TAYLOR—BURDEN 1970). The problem requires and is worthy of further extensive studies.

Arrest of leaf senescence by cytokinins has been explained on a basis of a hypothesis, supported by many experimental data, that cytokinins maintain a high rate of protein and RNA synthesis (cf. a review by OSBORNE 1965). As for bean cv. Pinto leaves, POZSÁR *et al.* (1967) have found that BA markedly promotes leucine and adenine incorporation into proteins and RNA, respectively; BA itself is also incorporated into Pinto bean leaf RNA (MATOLCSY *et al.* 1969). Promoted incorporation of the precursors into protein may genuinely reflect an effect of BA on the rate of protein synthesis, but it can also be explained on a basis of a hormone effect on amino acid uptake into the tissue or an effect on endogenous amino acid pool size resulting in isotope dilution. Some of these possibilities can be excluded by a study of the loss of radioactivity from the leaf tissue in which the protein fraction was pre-labelled. Recent studies of this type have revealed that cytokinins and some other growth regulators arrest yellowing by inhibition of protein breakdown (KURAISHI 1968, TAVARES *et al.* 1968, 1970, KNYPL 1969, 1970b, KNYPL—MAZURCZYK 1971, MIZRAHI *et al.* 1970). Although similar tests have not been performed with bean leaf tissue, it is quite possible that BA inhibits yellowing of the primary leaves held in darkness either by maintenance of protein synthesis (POZSÁR *et al.* 1967) or by arrest of proteolysis, or both. There is some evidence that cytokinins may accelerate protochlorophyll synthesis in darkness (Shlyk—Averina 1969, Shlyk *et al.* 1970); it is, however, improbable that that is the case in primary bean leaves in view of the facts that (1) light is required for the conversion of protochlorophyll to chlorophyll, and (2) BA initially inhibits chlorophyll synthesis (Jacoby—Dagan 1970).

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WHY WAS THE DECOMPOSITION OF LEAF PIGMENTS IN EXCISED LEAVES DELAYED BY BENZYLADENINE ONLY IN THE DARK?

o In the previous number of Acta Agronomica, under the title: "Effect of benzyladenine on the amount of leaf pigments in bean" Zatykó J. M. presented interesting experimental data tn the question whether chlorophyll decomposition could be hindered or delayed by synhetic cytokinins.

To study the physiological bases, the background of senescence is one of the most interesting problems of biology in our days. Of course, plant physiologists, breeders and growers are primarily interested in the problems of senescence in plants. Several years ago a very interesting summary written by MOTHES (1960) on this subject was published in the periodical Naturwissenschaften (*15*, 337, 1960); the author examined the process of senescence from the point of view of correlation between young and aged organs.

Paleness of leaves (chlorophyll decomposition) is considered to be a visible sign of senescence caused and induced by the insufficient activity of roots. Consequently, the life of old leaves can be prolonged by the activation of roots or elimination of competition (removal of younger organs). Old, yellow (but still living) leaves when sprayed with a 0.01—0.05 Mol solution of ammonium nitrate recover their green colour and begin to grow. This means that in a given case improvement in the nitrogen balance starts a process that may be called "rejuvenation".

By means of rooted leaf cuttings used it has been proved that the root is not only the organ of nutrient uptake but has also a role in the prolongation of life. Rooted leaf cuttings — unlike the rootless control — can be kept alive for a considerable time with their protein — and dry matter contents simultaneously increasing.

Zatykó's above mentioned paper describes experiments made similarly with rootless plants, excised bean cuttings, where the senescence of the experimental object was practically induced and hastened by the removal of roots.

Of the numerous parameters of senescence it was the decomposition of leaf pigments and its delay respectively, that formed the subject of the author's examination. He used benzyladenine — one of the most active synthetic cytokinins — as protective. His experimental results partly confirm the earlier findings, partly complete the special literature on this subject. It was only in the dark that benzyladenine hindered or delayed the decomposition of leaf pigments in the excised leaves, in the light no similar action was observed. This finding — as the author says — is "unexpected". Indeed, Osborne—McCalla's statement (1961) of kinetins exercising a protective effect on the decomposition of chlorophyll has been accepted by the literature (Fig. 1). It is not probable that this contradiction could be traced back to differences in the methods applied, though the methods of extraction used by the two authors were not identical, and they measured the pigment content in different ways.

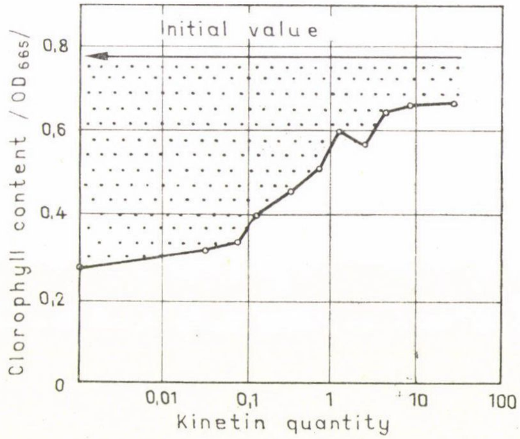

Fig. 1. Chlorophyll "protecting" effect of kinetin in green leaves. Chlorophyll content of discs excised from *Xanthium* leaves and floated for 48 hours on a solution containing kinetin was extracted with 80 per cent ethanol and measured at 665 nm. The initial amount of chlorophyll is indicated by the arrow (Osborne—McCalla 1961)

The difference found between chlorophyll-a and chlorophyll-b in the rate of decomposition is a very interesting result as it is consistent with the inhibiting effect on chlorophyll synthesis of another highly effective native hormone: gibberellin. In our experiments ("Gibberellinsäure und Chlorophyllgehalt des Blattes von *Phaseolus vulgaris* L." Planta (Berl.) *83*, 161—165, 1968) increasing concentrations of gibberellin were found to have a much more intensive inhibiting effect on the synthesis of chlorophyll-a than on that of chlorophyll-b (Fig. 2).

There is a striking similarity in the case of carotenoids and xanthophyll components too. Zatykó points out that neoxanthin is more resistant to photodestruction than the other components; in our experiments too, it was neoxanthin which was the kind of xanthophyll on which the most diverse concentrations of gibberellic acid had no effect (Fig. 3). The validity of the comparison would be increased if the experiments had been carried out with the same method. The author's attention should be called to the chlorophyll eluation, separation and determination methods employed by Hager—Meyer–Bertenrath (1962, 1966) as well as by Wintermans—Mots (1965), and widely used in chlorophyll pigment tests, by which the comparability of his further results can be increased too.

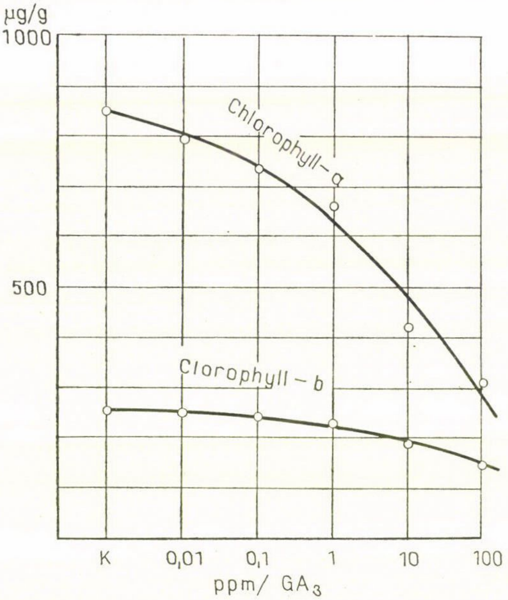

Fig. 2. **Effect of various concentrations of GA_3 on the amounts of chlorophyll-a and chlorophyll-b 10 days after the beginning of the experiments (Szalai 1968)**

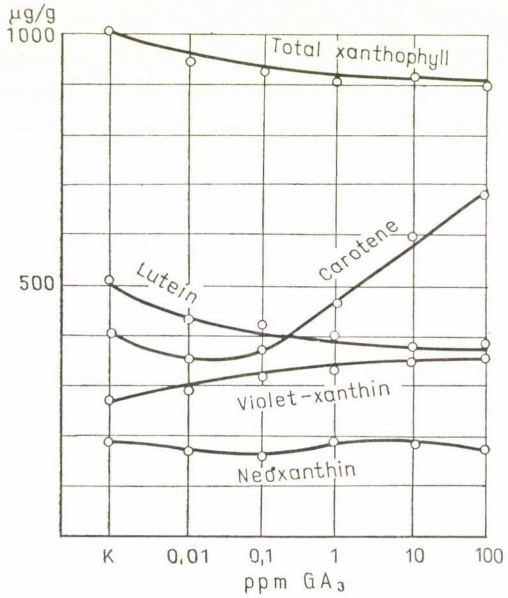

Fig. 3. **Effect of various** concentration of gibberellin on the amount of yellow pigments in the leaf (Szalai 1968)

The author's statements, as read in the discussion, are in a certain respect right, as it is highly probable that the survival and decomposition of pigments in senescing leaves depend on the intactness and denaturalization of chromoproteins, respectively. In order to make further advancement, the effect of benzyladenine and natural cytokinins (zeatins, triacanthin, etc.) on the synthesis of nucleic acids and proteins ought to be studied, as suggested by earlier observations (MOTHES 1960) on the mobilizing effect of cytokinins showing close correlations with the protein- and nucleic acid content as well as the age of leaves. On the other hand, it does not seem justified enough — at least not on the basis of the author's present experiment — to speak in this relation either of the protective effect of carotenoids or of the existence of a "feedback" mechanism. Finally, the not quite clear tables should be replaced by diagrams much more meaningful for the reader.

It is a good thing that investigations in this subject have started; we hope they will also continue. The problem would be more easily solved — or at least approached — if changes occurring in the course of natural senescence were also taken — as a control — into consideration; changes in the plastids — if not the whole sequence of senescence — followed with attention, namely the decomposition of proteins and the related changes in the amino acid level, the structural degeneration of chloroplast and ranule membrane, concentration of lipids into drops in which e.g. carotenoids are dissolved, etc.

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DECOMPOSITION OR DECREASE?

The paper by Dr. Zatykó contains some new information on the behaviour of carotenoids in detached leaves in the light and the dark, and the effects of a cytokinin. However, in relation to the contents, the paper is much too long. The second paragraph of the Introduction has no importance for the author's own work and can be deleted. Materials and Methods contain many details which I feel are not essential for the work. The Discussion either says that the author cannot contribute anything new to the problem, or simply repeats, perhaps in somewhat different words, what was said in the Results section. The only suggestion which is worth making is that about the protective action of carotenoids, although it must be added that the experimental evidence is still far from compelling. The statement about the "feed back" system seems to me, at this stage of the author's work, quite meaningless.

Altogether, I would recommend to condense the text of the paper to not more than one half. This can be done without any loss is interesting information. The documentation in four tables in also quite excessive. All pertinent information can be easily given in two tables. Giving values with two decimal points is also entirely pointless: the methods are not nearly that accurate!

A couple of individual points: The author is only measuring the content of the different pigments; he should therefore not speak of "decomposition" or similar processes, but of "loss" or "decrease". If differences are statistically not significant this means that there is no difference, and it is not permissible to say that there is.

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DOES BENZYLADENINE, WHEN IN DARKNESS, ALSO INHIBIT THE DESCTRUCTION OF THE CHLOROPLAST STRUCTURE?

Under normal conditions, that is in intact plants too, the composition and decomposition of the assimilation pigments, and their quantitative relations respectively, are — according to our present knowledge — in close correlation with the ultrastructure of the photosynthetic apparatus, and with the degree of its differentiation. In the case of a normal chloroplast development the process of the chlorophyll content rising to the proper level takes place simultaneously with the formation of the characteristic lamellar–granular structure. Similarly, the natural transformation of the chloroplast into chromoplast occurs parallel with the decomposition of the chlorophyll and the destruction of the lamellar structure. The above, as well as the biochemical data presented in the paper suggest that the pigment content decrease occurring in detached shoots was accompanied, in the experiments in question too, by the gradual destruction of the ultrastructure of the chloroplasts. Of course, this point can be cleared up only with electron microscopic examinations or — to some extent — with polaroscopic observations. If, however, it is really true — and we may well suppose so — it means that in the dark benzyladenine also inhibits the destruction of the chloroplast structure. Since chlorophyll decomposition, and with it probably the destruction of the structure too, takes place in darkness at a considerably higher rate than in light, and since — according to the author's experiments — it is only in darkness that benzyladenine is able to inhibit this process, it may be supposed that the whole process of destruction has a factor based on, and one independent of the absence of light. It is known that chlorophyll in its natural state is in close structural connection with the proteins, and in a certain sense with the lipoids too, thus it is probable that benzyladenine inhibits those processes occurring in the absence of light which lead to the breaking of connection between lipoprotein membranes and chlorophyll molecules, and to the destruction of the whole normal granular–lamellar structure.

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IS IT POSSIBLE TO INTERPRET COMPLICATED PHENOMENA WITHOUT STUDYING ITS SINGLE COMPONENTS?

Besides the induction of various other effects cytokinins are known to delay the decomposition of chlorophyll in excised senescent shoots and leaves. In the present paper data are given of phenomena which were observed when benzyladenine treated bean cuttings were exposed to light or darkness. Carotenoids were under investigation as well as chlorophylls.

Benzyladenine only reduced decomposition of chlorophyll in darkness but not in light. A constant ratio chlorophyll/carotenoids was maintained under these conditions as well as under continuous illumination. Carotenoids were scarcely affected by benzyladenine in light darkness, with the exception of neoxanthin.

Benzyladenine is thought to act via tRNA synthesis. Therefore it makes sense that photodecomposition of leaf pigments, the ability to photosynthetize new substances, the stability of chlorophyll–protein–carotenoid–complexes, and the enzymatic formation and decomposition of these complexes are influenced by benzyladenine in a different manner. It is scarcely possibly, however, to give an interpretation of phenomena based on a very complicated system without the knowledge of the single component. The experiments should therefore be the starting point for investigations on the protein (enzyme) and nucleic acid level.

Annotations:

1) The conversion of the experimental data to graphs would facilitate legibility.

2) The high standard deviations (?) of the experimental data should be discussed.

3) Hot water treatment before pigment extraction causes formation of pheophytines, isomeric and allomeric chlorophylls.

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IS THE LOWER PIGMENT LEVEL IN THE LEAVES OF ISOLATED SHOOTS CAUSED EXCLUSIVELY BY DECOMPOSITION, OR CAN THE DECREASE OF LIGHT DEPENDENT SYNTHESIS BE ALSO RESPONSIBLE FOR IT?

From a number of recently published works cytokinins are known to be able to normalize the metabolism of senescing leaves by retarding the decrease of nucleic acid-, protein- and chlorophyll levels characteristic of senescence. In senescing leaves decrease in the chlorophyll content runs largely parallel with changes in the protein- and nucleic acid contents of the leaves as well as of their capacity of carbon dioxide fixation, therefore chlorophyll loss is considered as a biochemical parameter of senescence and used for tests.

Considering the large number of publications dealing with the retarding effect exerted by the cytokinins — including benzyladenine — on the decomposition of chlorophyll, which mostly offer the same conclusions, the title of Zatykó's paper suggests a normal — not too promising — work. Still, the author was able to contribute to the subject by analysing the effect of benzyladenine not only on the chlorophyll content but on the carotenoid level too, and trying to establish causal relation between changes observed in the level of the two pigment groups. Investigations into changes occurring in the carotenoid level in the course of senescence as well as into the effect exerted by the cytokinins on the carotenoid content were — apart from a few exceptions (GASPAR—XHAUFFLAIRE 1968) — indeed completely neglected, although these pigments are remarkable due to the interesting features of their physiological role.

In addition to their role as energy transmitting accessory pigments, the physiological importance of carotenoids lies in the fact that through their own light absorption they protect — like a colour filter — the highly photolabile photosynthetic apparatus from photodestruction (FARKAS 1968), as shown e.g. by the high light sensitivity of carotenoid-free mutants.

In spite of the fact that many authors (VICKERY *et al.* 1937, PARTHIER *et al.* 1961, 1964; SUGIURA 1963, HOPKINSON 1966, etc.) referred to the retarding effect exerted by light on leaf senescence, relatively few publications deal with the question of interaction between light and cytokinins in relation with the changes occurring in the pigment level during senescence. That is why the author's efforts to study the effect of benzyladenine on leaf pigment content as a function of illumination is appreciated.

Observations and questions raised and proposals made in connection with the work are the following:

As to the method applied, it would have been better to carry out the extraction and peparation of the pigments in poor indifferent light or in darkness, without oxygen, thus preventing the pigments from being destroyed to some extent during the procedure, which may be a source of error.

In a work with a subject like this distinction should be made between natural and induced senescence, the latter being the case — through isolation of shoots or leaves (parts of leaves) — in the author's experiments as well. This way of inducing senescence is generally used; recent data suggest, however, that the two processes cannot be considered totally identical. LÁZÁR (1970) e.g. when studying the pattern of nucleic acid changes occurring in senescing leaves arrived unambiguously at this conclusion. Since similar comparisons have not been made so far concerning the leaf pigments, the problem is worth being paid attention to, all the more so as the question, whether the author's results can be applied to naturally senescing leaves, may be raised. After all, the real purpose of biological experimentation is to clarify and understand the in vivo physiological processes.

When continuing the work it would be worth while to find out whether the decrease of the pigment level in the leaves of isolated shoots is exclusively the result of decomposition, and to what extent the decrease of light dependent synthesis can be made responsible for it. Namely, in leaves isolated when young the protein- and chlorophyll synthesis was found to continue (SUGIURA 1963, PARISH 1968).

The author's suggestion that benzyladenine does not prevent the loss of chlorophyll in light is surprising and contradicts the known literary data (SUGIURA 1963, ENGELBRECHT 1964, GOLDTHWAITE—LAETSCH 1967, GASPAR—XHAUFFLAIRE 1968, FLETCHER 1969, PHILLIPS *et al.* 1969) according to which the cytokinins are efficient in retarding the decrease of chlorophyll level even in light (sometimes more intensively than in darkness). What explanation can the author offer for his divergent results? Would the employment of different experimental conditions and other cytokinins lead to the same conclusions? The question is raised because the influence exercised by the cytokinins on senescence is known to depend to a considerable extent on the intensity of light (GOLDTHWAITE—LAETSCH 1967, FLETCHER 1969, BACK—RICHMOND 1969), the method of isolation (LEOPOLD—KAWASE 1964), the age of isolated leaves (BACK—RICHMOND 1969), the applied cytokinin and its concentration (GASPAR—XHAUFFLAIRE 1968, BACK—RICHMOND 1969).

It is an interesting theory that in leaves of isolated shoots exposed to light — irrespective of the treatment — the unchanged chlorophyll/carotenoid ratio originating from decompositions of identical rate can be attributed to the protection of the carotenoids against photodestruction. This would practically mean that in light the level of chlorophylls in the leaf is determined by the actual carotenoid level, and that chlorophyll loss during senescence is due primarily to photodestruction. The latter is, however, inconsistent with the observation that chlorophyll level decreases more intensively in darkness (in shoots incubated in water the chlorophyll content fell to one-third in darkness, while only to one half in light, that is, illumination slowed down the disappearance of chlorophyll).

The "feedback" system assumed in relation with the protective role of the carotenoids requires a somewhat more detailed explanation.

The nature of light in the interaction between benzyladenine and light should similarly be clarified in the future: whether it is a photosynthetically active light that can be blocked by DMCU — as assumed on the basis of results obtained by PARTHIER *et al.* (1961, 1964) and GOLDTHWAITE—LAETSCH (1967) — or the interaction is a process mediated by phytochrome (SUGIURA 1963, TASSERON-DE JONG—VELDSTRA 1971).

As it can be seen Zatykó's work raises some actually and perspectivically interesting problems; this in itself is enough — not to mention its other merits — to draw attention to it.

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SHOULD THE PIGMENT CONTENT BE RELATED TO THE DRY WEIGHT OR THE LEAF AREA?

We are pleased to join in the discussion as we consider the work topical and the results obtained very valuable. Naturally we don't feel ourselves to be competent to discuss all the details, especially regarding the analytical part and the behaviour of the individual carotenoids. Therefore we must be content with only the discussion of the differences between the individual treatments and with that of their connections. Mainly Tables 2 and 4 (Pigment content of dry materials) served as the basis for discussion.

1) Treatments in dark:

a) The chlorophyll behaviour corresponds to the status verified again and again by the examinations of Richmond and Land, Mothes and co-workers and other authors, respectively. The decomposition rate might have been expressed even more clearly if the pigment content had not been related to the fresh or dry weight also changing in the dark. The considerable change in fresh and dry weight rate can be demonstrated by the rate of one of the pigments, e.g. chlorophyll a, related to g fresh weight as well as of the chlorophyll a content related to g dry weight. Reflecting the relationship between fresh and dry weight, this is modified from (7333 μg/954 μg =) 7.7 (basic material) to 10.0 (water treatment) or to 12.1 (BA treatment). This difference must be taken into consideration when comparing the fresh and dry matter contents. A clearer picture could have been formed of the behaviour of the pigments against the basic materials if the pigment content had been related to leaf area as it remained unchanged during the experiment.

b) Carotenoids proved to be much more resistant than chlorophyll. This result was placed on a broader base by the thin separation of the xantophylls. The fact that the control values exceed the basic ones is only a consequence of the choosing of the base of comparison. Supposing a decrease in dry matter of around 10% within 5 to 8 days, the quantity of carotenoids also decreases.

c) BA seemed to accelerate the decomposition of the carotenoids as compared to the watered control. In the case of BA treatment after 5 to 8 days only 86.9% of the total carotenoids were present as compared to the 101.6% of the control. However, the choosing of the base of comparison here too, must be taken into consideration. During the experiment the dry matter can change to various degrees. Its amount is supposed to be highly decreased when placing the offshoot in water, due not only to respiration, but also the outlet on the stems left on the leaves. Supposedly its retention strength was increased to such an extent by the BA-spraying of laminae, that due to the force of attraction (Mothes 1960) even a transport of material takes place from the stems to the leaves, so their dry matter content per leaf or leaf area did not necessarily decrease. In this case 86.9% of the total carotenoids treated with BA would correspond to the actual decomposition, while in the case of 101.6% of the watered control the high decrease in dry matter (over 10%) should be taken into consideration. For this reason, from the data available it cannot be concluded with absolute certainty whether the BA would accelerate the decomposition of the carotenoids (or would protect them to a lesser extent). This conclusion cannot be made without the data concerning the dry matter loss per determined unit of leaf area.

Independently of this consideration the new result of the paper is reliable, i.e. irrespectively of the manner of treatment the 2.2 xantophyll/carotene ratio is constant as well as the 2.5 chlorophyll a/b ratio.

2) In constant light:

a) According to the expectations in this case the decomposition processes are inhibited. Using a watered control the author succeeded in light, during double time (10 to 15 days) to get the chlorophyll content related to the dry matter equivalent to that achieved in the dark treatment of 5 to 8 days, i.e. 45 to 50% of the basic material related to dry matter. Chlorophyll decomposition is not delayed by BA in light. The reason for this may either be that BA is sensitive to light, or that there supposedly exist other (not directed) phytohormone systems similar to the cytokinins.

According to the recent conceptions the decomposition processes take place in the following way: RN-ase activity increases in excised leaves especially in the dark, the protein "turnover" is affected, and the protein synthesis is surpassed by the protein decomposition. During the decomposition of the plastides and in the case of their structural desorganization the pigments become free. The chlorophylls are decomposed at once, this is why a close connection exists between protein and chlorophyll decomposition. At this stage the carotenoids are more resistant, their decomposition takes place more slowly. Cytokinins like BA inhibit RN-ase activity (or increase the resistance of RNA to RN-ase), so the protein "turnover" is not damaged and the proteins, plastides, plastid pigments, etc. remain unchanged. On this basis the protein is in a dominant position, the decomposition of which causes the decomposition of numerous other materials (for example that of linoleic acids too; MATAR 1971). Unfortunately the protein content was not determined.

b) Mr. Zatykó sees a comparison between chlorophyll and carotene decomposition in light, in connection with a "protective effect of carotenoids against photodestruction". In reality a high decomposition of the carotenoids can be experienced in light, parallelly with the relatively good stability of chlorophyll, which must have decomposed to a higher extent during the long (10 to 15 days) period as compared to the 5 to 8 days treatment in dark. However, it cannot be concluded from the data available, whether the connection between the two phenomena: the fast carotene decomposition and the relatively low chlorophyll loss, was casual. If we agree with the opinion about the process of decomposition and the central position of the proteins described above (under 2a), it may be supposed that the chlorophyll is again in close correlation with the protein, and that the protein decomposition is primary. In this case the "protective effect" of the carotenoids may have a deeper cause referring first of all to the "turnover" of the proteins, cca in the sense mentioned above, for which nowadays there are no data available exact in all respects. For this reason too, it would be very necessary to obtain data referring to the protein content.

3) The present experiments gave us a whole range of valuable new findings and raised, as usual, a lot of new problems. Further experiments are needed especially on the direction mechanism in light, to make the results published satisfyingly explainable. Therefore the question is worth a further work.

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ARE THE PIGMENT DECOMPOSING EFFECTS OF ROOT REMOVAL AND DARKNESS BASED ON DIFFERENT MECHANISMS?

Zatykó's paper provides further information on the action mechanism of benzyladenine. The difference in behaviour pointed out between shoots kept in darkness and those kept in light is especially interesting, and suggests that the pigment decomposing effects of root removal and darkness are based on different mechanisms.

It is known, that in the dark, pigments decompose even in rooted shoots, which is more pronounced in the chlorophyll than in the carotene content. In shoots excised and kept in the dark the two pigment decomposing effects probably sum up. This is supported by the fact that the visible signs of pigment decomposition appear much earlier in shoots kept in darkness than in those kept in light. Benzyladenine only seems to be able to compensate for pigment decompositions occurring as a result of darkening.

The author's statements concerning the decomposition of neoxanthine are based on differences too small to be sufficiently convincing.

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WHY DID CYTOKININ NOT RETARD SENESCENCE IN SHOOTS MAINTAINED IN CONTINUOUS LIGHT?

The reported work seems to represent a careful and accurate study. There have been many similar reports on the effect of growth regulators on leaf senescence, but this one contains some original observations on the effect of a cytokinin on carotenoids.

The fact that the cytokinin did not retard senescence in shoots maintained in continuous light merits greater attention in the discussion. I presume that photo-oxidation cancels out the cytokinin effect. Attention should be called to other reports in the literature where senescing tissue was maintained in the light. That of Leopold—Kawase (Amer. J. Bot. 51: 294; 1961) is an example.

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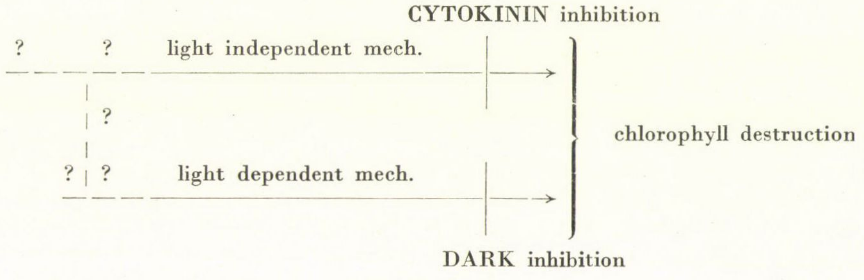
ON TWO POSSIBLE MECHANISMS OF THE DESTRUCTION OF LEAF PIGMENTS

In his paper, Zatykó (1971) publishes very interesting data on the effect of a cytokinin benzyladenine (BA) on the pigment content of isolated bean leaves. His observation that the destruction of chlorophylls can be inhibited by BA only in the dark is remarkable. In the experiments mentioned above the destruction of xanthophylls and carotenes cannot be inhibited by BA.

The cytokinin effect has an important role in the processes of differentiation and morphogenesis (Skoog—Miller 1957, Doerschug—Miller 1967). The cytokinins significantly influence the metabolism of nucleic acids and proteins which results in changes in plant growth and development. Isolating the leaves from the intact plant increases their senescence. On the other hand, the processes of senescence can be inhibited by cytokinins (Richmond—Lang 1957). In leaves during senescence the destruction of proteins and chlorophylls can be experienced and the cytokinins can reduce their destruction (Osborne 1964). The process of senescence, after all, represents the final stage of ontogenesis; that is in this case the cytokinins also influence the processes of development. The metabolism and senescence of isolated leaves may be different in light and in darkness. The effect of light on the photosynthetic pigments and on their destruction may be direct or indirect. The effect of light depends on a number of factors and it is not in connection with the morphogenetic processes under every circumstance in the case of pigment destruction.

It has been established that the albino gene (w_3) of maize has a significant morphogenetic effect. Seedlings of the normal and albino mutant maize germinated in the dark have a different ratio of coleoptile and mesocotyl length (K/M). In spite of different morphological characters (the different length ratio of coleoptile and mesocotyl) a similar leaf pigment content could be measured in the dark (Kovács 1962). In light the change of the morphological character, the K/M ratio shows a similar tendency but the pigment content of the normal seedlings is increased while that of the albino mutants is reduced by photo-destruction. These experiments clearly show that the gene mutation on the w_3 locus resulted in a change essentially of the morphogenetic processes secondarily influencing the stability of the leaf pigments in light (Kovács 1962, 1963).

On the basis of the facts mentioned above, we can assume the existence of two different mechanisms for the destruction of the leaf pigments in isolated plant parts, e.g. in leaves. One of the mechanisms operates independently from light and it is sensitive to the cytokinin effect. The other mechanism is a light dependent process and it may be insensitive to cytokinins. In light both of the mechanisms may operate but the more direct destruction effect of the light process may hide the operation of the light independent mechanism. Thus the cytokinin effect cannot be detected. The relationship between the two mechanisms is demonstrated in the following figure:



The light independent destruction process may be a component of the normal regulation mechanisms of the developmental processes influencing cell differentiation as well as the differentiation of cell organelles including chloroplasts, and its effect is more general. Presumably, the light dependent process is a type of photo-destruction mechanism where the direct or indirect effect of light leads to the destruction of the pigments (e.g. chlorophylls).

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IS CHLOROPHYLL MORE SUSCEPTIBLE TO SENESCENCE THAN THE CAROTENOIDS?

Since our own work on senescence of leaves has concerned only the chlorophyll and protein components, this work demonstrating changes in carotenoids is of great interest. The author is to be congratulated on having collected such extensive and apparently reliable data.

Put simply — in the dark the chlorophyll drops almost to one third of the initial value, but the yellow pigments drop only some 20%; benzyladenine partially arrests the drop in chlorophyll (holding it to 50% of the initial) but does not affect the small drop in yellow pigments. In light, on the other hand, the chlorophyll and carotenoids both drop to 50% of the initial value and benzyladenine does not affect either one. There may be several explanations for this:

a) The chlorophyll value after the 10—15 days in light was almost exactly the same (about 50%) as in darkness with benzyladenine; this could be interpreted as meaning that this value is the most that 5 ppm benzyladenine can produce. Higher concentrations of the cytokinin should be tried.

b) Benzyladenine does not significantly affect the yellow pigments in either light or darkness. This merely shows that the cytokinin effect is exerted on the chlorophyll and not on the carotenoids. Since in the dark the concentrations of the two types of pigments can vary independently, there is no reason to invoke a feedback relationship between them.

c) Our work has shown (Plant Physiology 46, 212—220, 1970, and Proc. Canberra Conference on Plant Growth Substances, 1971 in press) that proteolysis dominates the senescence process, at least in the dark. It may be, therefore, that chlorophyll is more susceptible than the carotenoids because it is bound to protein in the plastid and owes its stability to this binding, while the carotenoids are either not so bound or else are equally stable whether bound or not.

In any event I feel one should concentrate on the actual (or percentage) values and not on the ratios, which can be misleading. The data should be given with less decimals — three significant figures are probably all that the methods justify, and do not need to be given both for dry weight and for fresh weight; this would make them easier to review and would bring out the changes in absolute values.

As a detail, the realization that chlorophyll is combined with protein in the plastid is not "recent"; it dates back to the 1940's.

Work of this type should be continued and will eventually shed light on a great mystery of plant physiology — the phenomenon of senescence.

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DO KINETIN-LIKE COMPOUNDS EXERT THEIR INFLUENCE THROUGH PROTEIN SYNTHESIS IN EVERY CASE?

The effect of kinetin on the pigment level of detached leaves is well known; but there is no information on the effect of benzyladenine, and its action on the metabolism of carotenoids. For this reason the author's experiments may arouse interest. Concerning the experiments I should like to make a few remarks:

1. On the basis of the author's data (Tables 1 and 3) the decrease in the carotenoid level due to detachment takes place to the same extent in light as in darkness, and benzyladenine is not able to inhibit this process. This fact suggests that the kinetin-like compounds cannot in every case exert their influence through protein synthesis. The antagonism existing between kinetin and some amino acids (SHIBAOKA—THIMANN 1970) also points to the possibility that only a decreased protein decomposition rather than a general favourable effect exerted on the protein synthesis can be reckoned with. The effect of kinetin-like compounds on various plants — as mentioned by the author — also seems to confirm that in the case of kinetin-like compounds a certain degree of specificity must be taken in consideration.

2. The different light-resistance of chlorophylls and carotenoids, as well as the low light intensity used in the experiments are factors which by themselves are enough to make the existence of photodestruction assumed by the author questionable. The different effects that the kinetin-like compounds exert on the chlorophyll- and carotenoid levels of leaves are more probably due to the existence of light-dependent protein synthesis processes and the influence of kinetin-like compounds — including benzyladenine used in the author's experiments — on these processes.

3. In our investigations made with wheat leaves the greening processes in the etiolated leaves were normal even 96 hours after detachment although during the same period the chlorophyll decomposition in detached green leaves kept in darkness was nearly completed. Etiolated leaves generally lost their "greening ability" within 6—7 days after detachment, and this process could not be retarded either by carbohydrates or kinetin application.

4. Senescence induced by detachment caused metabolical changes of different character in light and of different character in darkness (KISBÁN *et al.* 1964, NGUYEN VAN UYEN 1971). The senescence retarding effect of kinetin-like compounds developed mainly under dark conditions. Taking the light dependent enzyme synthesis indicated in bean leaves (BRADBEER 1969) as a basis, and taking the stabilizing effect of light on chlorophylls into consideration the influence of kinetin-like compounds should at all events be discussed making a distinction between light-dependent and light-independent protein syntheses.

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DOES BENZYLADENINE HAVE ANY EFFECT ON SHOOT LEAVES KEPT IN THE LIGHT?

In recent years great attention has been paid to the study of the physiological role of cytokinins. From the time when kinetin (6-furfuryl aminopurine) was obtained and its chemical characteristics determined, there has been a growing stream of information in this field every year. This phenomenon can be explained by its theoretical importance and perspectives for practice as well. There are numerous data available on the positive influence of kinins on plant metabolism and, in some cases its high effectivity is determined in connection with the synthetical processes (protein synthesis, etc.).

In many respects the latter evidently determines the defensive effect of kinins in the processes connected with structural destruction: increasing the resistance to ray injuries, the inhibition of senescence of isolated shoots, etc.

It has just been determined that the effect of cytokinins appears to be significantly stronger on the isolated plant organs having no connection with the root system, though a kinin effect of similar extent can be observed on the leaves of intact plants when nitrogen is eliminated from the nutritive substances (Kulaeva 1962).

In his paper J. M. Zatykó investigated the effect of one of the kinetin analogues — benzyladenine — on the senescence of isolated bean shoots.

A great number of kinetin analogues have been obtained up till the present time using various replacers in the purine cycle which have a similar plant physiological effect as kinetin. According to J. M. Zatykó's paper benzyladenine also has an inhibiting effect on the senescence of isolated bean shoots kept in the dark (5—8 days).

The author found that chlorophyll degradation decreased in these leaves compared to the leaves treated with distilled water which is in agreement with the literary data.

However, the results on the effect of benzyladenine on shoot leaves kept in the light (10—15 days) do not agree with the data obtained by numerous authors investigating kinetin derivates, among them benzyladenine. Namely according to the author the decrease in chlorophyll content after excising the shoots and keeping them in the light was equivalent both in the shoots treated with benzyladenine and in those treated with distilled water.

In numerous works (Mothes 1960, Kulaeva 1962) it was clearly shown that cytokinins are not only capable of inhibiting the senescence of the isolated shoots but can even cause their rejuvenation. Kulaeva (1962) showed that kinetin has an effect on chlorophyll synthesis in yellowed shoots.

There are also some data available on benzyladenine. It had a stimulating effect on chlorophyll synthesis in the isolated cotyledons of cucumber and pumpkin kept in the light (Knypl 1970/a, Knypl 1970/b).

A large amount of experimental material has also been collected in connection with the action mechanism of cytokinins on senescent cells. As it was shown by cytological investigations, in senescent cells, cytokinin has an accelerating effect on the structure-restoring processes of nuclei, nucleoli, mytochondria and chloroplasts, respectively.

Cytokinins, in part, are capable of preventing the membrane injuries of the chloroplasts, caused by X-ray (SVESNIKOVA 1970).

From the point of view of understanding the action mechanism of kinins on senescent cells, strict attention should be paid to the work of ADEDIPE (1970); benzyladenine is shown by him to decrease the diffusion of assimilates from bean leaves. The accumulation of assimilates must, no doubt have an accelerating effect on chlorophyll synthesis.

However, the author does not attempt to evaluate the facts determined by him as a positive effect of benzyladenine on senescent shoots kept in the dark, or as an uneffictivity of the latter when keeping the shoots in the light. In this case the absence of benzyladenine response cannot be said to be justified because in this work only one of the benzyladenine concentrations was used.

The author does not justify the choosing of the concentration of a not sufficiently studied representative of the cytokinin group.

It is not excluded that by the using of other concentrations it would be possible to observe a benzyladenine effect similar to that in the works cited above.

Great attention was paid by the author to carotenoids. From the wide Table in the work it is evident that the benzyladenine treatment of bean leaves did not inhibit the carotenoid decrease in light, in contrast with the data of XHAUFFLAIRE (1968). The author explains this contradiction with the "peculiar behaviour of neoxanthin"; its content is slightly decreased in the benzyladenine treatment.

The equilibrium of the $\frac{\text{chlorophyll}}{\text{carotenoids}}$ ratio when treated with benzyladenine and with distilled water is explained by the author as the effect of the defensive action of carotenoids in pigment photodestruction processes. This conception, however, does not seem to be satisfactory enough.

In conclusion it must be mentioned that the material collected by Zatykó is in fact worthy of scientific attention. Unfortunately, however, it has not been undertaken to a detailed analysis.

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DOES BENZYLADENINE HAVE THE SAME INFLUENCE ON THE FORMATION OF CHLOROPLAST LAMELLAE IN THE LEAVES OF SHOOTS KEPT IN DARKNESS AS IT HAS IN GERMINATING SEEDS?

The author's statements found in the results and discussion of his experiments are surprising from a physiological point of view as well. He points out that under the influence of benzyladenine treatments chlorophyll decomposition is inhibited — and to a considerable extent — in excised bean shoots kept in darkness, while in light it takes place in the same way as in the control plants but at a much higher rate.

The results are considered unexpected both from a plant organizational and a cytological aspect. According to our knowledge no cytological ultrastructure studies have been carried out under such experimental conditions.

The experiments showed that benzyladenine (50—100 mg/l) applied during germination increased the chlorophyll content of leaves to a great extent. According to the observations made with an electron microscope the number of lamellae increased in the chloroplasts. In the course of observations made on root organization we, too, found that not only the leaves but also the light green hypocotyl became fierce green, which indicated an intensive increase in the chlorophyll content as a reaction to benzyladenine treatments. Thus, benzyladenine has a great influence on chlorophyll formation, if this influence is exercised from the beginning of germination.

According to the author's investigations, without benzyladenine treatments the leaves of shoots kept in darkness lost their chlorophyll content within 5—8 days, which is sooner than when kept in light. From a plant organizational approach of the problem it may be supposed that in dark the etiolation processes occur in the structure of chloroplasts, the vesicula and lamellae begin to decompose, and during this process certain stages may be similar to the chloroplast structure developing during germination. If benzyladenine is applied at this stage, it enters the process of decomposition with the same effect as it has during germination, with the difference that here it prevents the chloroplasts from further destruction and occasionally results in the decomposed lamellae being replaced by new ones.

In this respect this problem would require two kinds of ultrastructural investigations: on one hand, it is to be found out what the development stages and structures during germination are at which formation of additional lamellae begins; on the other hand, with the method of investigation used by the author, what the initial signs of chloroplast decomposition are, and what structural peculiarities can be observed under the influence of benzyladenine, when chlorophyll decomposition stops.

With a closer approach to the question in view it would be worth-while to make benzyladenine treatments in dark in more than one variation, so as to render the evaluation of the effects of treatments more reliable.

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CAN THE "FEEDBACK" SYSTEM ENSURING THE BALANCE OF PIGMENTS BE CONSIDERED AS PROVED?

The paper deals with a synthetic hormone which exceeds the cytokinins in its juvenility maintaining and rejuvenating effect, and is thus highly important not only from a theoretical but also from a practical point of view.

The methods applied meet the requirements of the experiment. Reliability of data obtained is guaranteed by several repetitions made and a large number of parallel samples. However, the precision of the values could have been increased if the experiments had been carried out in a nitrogen atmosphere.

The language of the paper is clear and unambiguous, its reasoning is logical. Interpretation of the data is not too lengthy, the author endeavours to elucidate the most important correlations. Data on the peculiar behaviour of neoxanthin are considered especially valuable.

The paper is not confined to a registration-like static valuation of results. The author tries to find correlations in the decomposition of pigments. He points out that in leaves kept permanently illuminated the rates of chlorophyll- and carotenoid decomposition are parallel, and the phenomenon is in connection with the protective effect exerted by the carotenoids against photodestruction. This statement — though not novel in principle — is interesting because it proves the results found with carotenoidless mutants. As for the "feedback'- system ensuring the balance of pigments it is probable but not proved.

To sum up, the paper is an important work concerning both its results and concept, which contributes new data to our knowledge of plant physiology.

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DO CYTOKININS HAVE A PHOTOPERIODICAL ACTIVITY WHICH SUBSTITUTES ILLUMINATION AS WELL?

The author proved experimentally that in isolated shoots kept in darkness benzyladenine treatments inhibited the decomposition of chlorophyll while were ineffective in light.

This result suggests that synthetic benzyladenine displays an illumination substituting effectivity. In order to prove this supposition it would be right to compare the benzyladenine treatments with controls kept under long- and short day illumination (of 8 and 14 hours duration respectively), as well as only 1, 2 and 3 hours illumination, to the dark control. In this way the light substituting effectivity (biological effectivity substituting illumination) of benzyladenine could be experimentally valuated. On the other hand, the isolated shoots ought to be compared with rooted plants, or endogenous cytokinin extracts isolated from roots, since the purin and pyrimidin bases are first of all produced in the root. Endogenous cytokinin is presumably also produced in roots of plants kept in darkness, however, it is, quantitatively different from the cytokinin level of plants grown in natural light or under constant illumination. Under natural conditions the endogenous cytokinins are carried by xylem transport into the shoot apex or juvenile leaves; this model should also be compared with the leaf treatments, where parenchymatic translocation exists. Beside studying chlorophyll decomposition intensively in experiments, attention should also be paid to whether the synthetic cytokinins induce leaf pigment formation in etiolated organisms and under the influence of short illuminations of different wave length, after the preventive treatments. It is highly probable that the intensity of protein synthesis in leaves and the ratio of the protein fractions also have a part in inhibiting or retarding the chlorophyll decomposition in darkness when exogenous cytokinin is applied. In our opinion it would be right to prove the correlation by way of an experiment too in connection with the phenomenon studied. This supposition is confirmed in particular by the experimental result which shows that the cytokinins not only increase the intensity of protein synthesis, primarily by stimulating the nucleic acid synthesis, but also retard the decomposition of protein through a so far unknown mechanism, and as a result of this a decrease in the enzyme activity which, in fact, causes the inhibition of senescence.

Finally, in relation to the problem raised in the title, we should like to emphasize the biological effectivity of kinetin — the other well-known synthetic cytokinin — in influencing photoperiodicalness. According to our unpublished experimental results (Király—Pozsár—Sági: Induction of flowering by kinetin in a long-day plant under short-day conditions) *Nicotiana tabacum* (L.) var. Xanthi, which had not flowered under short-day conditions, when treated daily with kinetin (50 ppm) for three weeks, flowered in the same way as long-day plants kept in natural light for 8 hours and under a supplementing artificial illumination (5000 and 1500 lux respectively) for a further 8 hours. The light substituting-, photoperiodically inductive effect of kinetin is realized under short-day conditions probably either through the synthesis of endogenous gibberellins or through the formation of other biological effective compounds.

With the above taken in consideration, the biological effectivity of cytokinins in substituting light should be further studied, not only with chlorophyll level stabilization in view, but also in relation to photoperiodicalness, especially in the limiting marginal cases, in connection with the correlation of expositional doses, spectral range playing a role in induction and leaf pigment ratios.

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CHRONICA

MÁTYÁS MOHÁCSY

1881–1970

Mátyás Mohácsy the respected and much loved teacher of Hungarian horticulturists died on April 6th, 1970. He devoted all his life to developing Hungarian horticulture; without his work it could hardly have shown such a rapid progress as is taking place under our very eyes.

He was born on March 12th, 1881 at Békéscsaba in a family of seven children. His father was a tanner who had all his children educated. Mátyás Mohácsy got acquainted early with the elements of horticulture while helping his father — an expert of grafting and budding — in carrying out such work for the local farmers.

After the fourth class of the grammar school he went to study at the Horticultural School in Budapest, in 1897. Dezső Angyal the excellent pomologist and director of the School as well as his fellow teachers possessed thorough theoretical and practical knowledge. The curriculum was based on biology and practical problems were built on this basis.

Mátyás Mohácsy obtained his final certificate in 1900. He had an irrepressible desire to learn the advanced agrotechnics and methods of the western countries and introduce them in Hungary.

His first journey abroad was to Stuttgart, Germany in 1901 where he worked for a year on a farm in the fruit tree nursery, wall-tree garden and packing unit. It was there that he got acquainted with the then new, highly important pruning methods, with the modern packing and storing methods, and it was there, too, he realized that Hungary could produce excellent quality fruit, if the way of packing and transporting fruit for export purposes were known. He also acquired a thorough knowledge of the German language during that year.

His next trip was to Basel in 1902 where he worked at a landscape architecture firm. In the same year he was called up for a year of military service which he completed at Temesvár. In January 1904 he went to London and was employed in a forcing unit in one of the suburbs; he became familiar with both that branch of horticulture in London and acquired a knowledge of the language, too. He was especially interested in Sander's Orchid farm.

The next place he went to was France. First he worked at Versailles where he learnt ornamental growing, got acquainted with the methods of propagation, the oecological requirements of plants, the procedures of plant protection, the right way of packing, and with marketing; in addition at the horticultural school of Versailles he also studied orchards, especially the various forms of wall-trees. At the end of 1904 he went to Paris and was employed in one of the palm-houses of the Jardin des Plantes, but also studied parks, nurseries, orchards, wall-trees and household gardens, and in the meantime learnt French, too.

He attended public lectures at the Sorbonne and under their influence went to Nancy to V. Lemoine who employed him during 1905 and 1906. There he learned the methods of crossing and dealt with the question of origin and inheritance. It was there he realized that horticulture and varietal problems are inseparable.

In 1906 he was employed at an ornamental forcing unit in Berlin where his continued professional education was also provided for. He had an opportunity to visit the Academy of Horticulture at Dahlem where he became aware of the necessity of a close connection between education and research work.

It was there he was notified that the Ministry wanted to send him to the United States, but first he had to spend a year at home. He went home and worked for a year —from April let, 1907 to May 7th, 1908 — at Torda as the leader of horticultural practice at the Horticultural Worker's Training School. So far he had travelled at private expense, but now his travel reports attracted attention. In 1908 he was sent to the United States at public expense to study horticultural farms and get acquainted with fruit marketing and plant protection.

First he went to Kansas to a fruit producing farm where he had opportunity to learn planting, mass production, propagation material production, soil cultivation, plantation protection, harvesting, grading, packing, transporting as well as mechanization of every possible process in practice, too, and saw how production could and should be adapted to the consumers' demands quickly and flexibly. It was there he met the most up-to-date packing method of that time — in the so called "Oregon boxes", mechanical spraying, in dry regions flooding, in Weatsonville a highly efficient fruit drying installation, examplary farm management and accounting system.

He succeeded in visiting Luther Burbank, the world-famous breeder in Santa Rosa, near San Francisco who showed him some of his breeding methods.

He came home in November, 1911. His study tour lasted one and a half years longer than was originally planned but was worth-while, as Mátyás Mohácsy returned with a wide knowledge and experience which the whole country could make use of.

After his return he was appointed leader of the fruit growing unit of the Horticultural School that had made great progress in the meantime. At Budaörs he built up the fruit drier he had seen at Weatsonville and held demonstrations of fruit grading and -packing there.

In 1913 he married Margit Gubicza who proved a faithful and understanding companion throughout his life.

On the occasion of a large-scale fruit exhibition Mátyás Mohácsy attracted general interest with his fruits packed with the up-to-date method learnt in America. He was then assigned to central service and had opportunity to travel about the country and impart all the knowledge he had acquired abroad.

It was then that he was appointed to a directorship at the Horticultural Worker's State Training School at Nagybocskó, and his first child Margit was born.

When the first world war broke out he was called up for military service, and already in November 1914 taken prisoner of war and sent to the camp of Krasnoyarsk. In the camp he compiled two books on the basis of results and experiences of his travels abroad: one of them on the practice of fruit growing, the other on horticultural farm management. In 1919 he was given the possibility of organizing a horticultural unit in the camp mainly with seedling production and vegetable growing, which provided the necessary financial means for him to go home.

In June 1920 he arrived at Zirc (a small country town in Transdanubia) where he met his family. There he was visited by one of the teachers of the Horticultural School who invited him to be the teacher of fruit growing. The new director Béla Rerrich informed him of the major problems: to supply foreign markets — in addition to the home market — with horticultural products, and grant the horticultural students a high level education.

Mátyás Mohácsy began work with great enthusiasm. His lectures — and papers, too — were characterized by careful preparation, good structure and a precise and clear delivery. In his scientific work he adopted the principle of drawing conclusions on the basis of wide experiences only.

After his return he spent five years in compiling — with the help of his best students — the material of his former director Dezső Angyal in a book published in four volumes in 1925—26. In the meantime he was given lodging first at the Institute of Ampelology, then, at last, in 1925 an official residence at the Horticultural School. This removed all difficulties concerning his family which by that time had increased by one member: his son Mátyás. With his personal problems thus solved he could devote more energy to his work.

In spite of excellent natural conditions and great possibilities, fruit growing in Hungary did not reach the level it could have done. Considerable proportions of fruit trees were grown in private gardens, there were, also, scattered orchards, but orchards producing the quantity and quality necessary for meeting the export requirements could not be spoken of at that time — except some large estates and model state orchards. Mátyás Mohácsy realized that horticultural production could only be improved by establishing mechanized and well equipped commodity producing large-scale orchards. His book "Gyakorlati gyümölcstermesztés" (The practice of fruit growing) published in 1922 which ran into five editions served this purpose.

The stable framework of Hungarian horticultural production gradually developed. With his collaborators Mátyás Mohácsy set up fruit growing districts and determined their optimum fruit sorts, thus preparing a large-scale fruit production which could already rely on safe market research data. E.g. the county of Szabolcs-Szatmár marked out as a winter apple district greatly corresponded to the aspects of plant geography.

A National Committee of Pomology determining the sorts suitable for large-scale production as well as the volume of production was also established.

Another — already prepared — work of Mohácsy: "Kertészeti üzemtan, jövedelem és értékszámítás" (Horticultural management, income and value calculation) as well as several papers on fruit growing were published in those years.

Then he went on study tours again to Germany, France, Holland, Austria, Turkey and Italy.

In 1928 his third child, Maria was born.

In 1930 a decree was issued declaring the Horticultural School to be a secondary school. The decree evoked general indignation, as it was issued at a time when the School began to play the role of a higher educational institution due to its economic and scientific importance.

In the same year Mátyás Mohácsy was appointed director of the School and began to play a leading role in promoting the cause of horticulture equally in education, research and organization. In 1927 horticultural production was performed only on some 1.2 per cent of the total area under cultivation, and horticultural products amounted to less than 2 per cent of the total export. This situation had to be and was changed by increasing the production of horticulture and entering into competition on foreign markets.

Another aim was to have the importance of the School acknowledged. Mátyás Mohácsy persuaded the teachers to obtain degrees at various higher educational institutions in order to prove the scientific importance of the School. Also he edited a yearbook entitled "Tanintézet Közleményei" (Bulletin of the School), containing scientific papers within the domain of the institution.

Large-scale mechanized orchards were being planted one after the other; he himself established a model orchard in county Szabolcs which later proved to be an excellent experimental area.

In those years he was visited by a terrible blow: his daughter Margit died a couple of days before her wedding.

In the thirties he was elected leader of various associations: the Hungarian Society of Tree Nursery, the National Association of Fruit Growers, the National Committee for Judging Plant Novelties, the National Board of Fruit Growing Experts, the National Committee of Pomology, the Society of Certified Horticulturists.

In the first 5 years of his directorship the number of fruit trees redoubled in the country: by 1938 Hungary was not dependent on fruit import moreover, fruit export become profitable.

At last, in 1939, the institution was reorganized into an independent Academy of Horticulture with Budapest as centre and Mohácsy as director. Three years later the Academy was completed with the branches of viticulture and oecology and became College of Horticulture and Viticulture. Mohácsy was promoted to a professorship and elected as dean for 1943—44.

Between December 15, 1944 and February 11, 1945 the College was hit by more than 3000 bombs destroying everything built up during so many years and even causing losses in human life.

However, work soon started under the leadership of Mohácsy. In 1945 the College fusioned with the other agricultural higher educational institutions in the framework of the University of Agricultural Sciences and continued to function as an independent Faculty of Horticulture and Viticulture. Mohácsy was appointed to a professorship and his students presented him with Vol. 10. of the Bulletin compiled by him which was the first publication of its kind after the war.

In the following years Mohácsy took part in developing the large-scale farms.

In 1948—49 he was elected rector of the University of Agricultural Sciences, and in 1949 was awarded the Kossuth prize. In the same year his old desire: the unity of education and research came true with the Horticultural Research Institute established at Budatétény, where research work was carried on under the guidance of the department leaders of the University.

At the end of 1949 he retired: in the same year his son Mátyás was appointed teacher at the Department of Fruit Growing.

The following years were spent in undisturbed creative work. He owned an orchard at Sasad, near Budapest, where he regularly went to work, to do experiments, make investigations together with his son, or — in bad weather — write down the results and experiences of a long life. 33 works originated from this period, most of them with co-authors — his son among others.

But the quietly living scientist was not forgotten. In 1952 he became candidate; in 1954 was decorated with the Order of the Red Banner; in 1957 declared doctor of agricultural sciences of fifty years standing by the Hungarian Academy of Sciences.

In 1958 Mohácsy suffered a great loss: his son Mátyás, father of two little children suddenly fell ill and died. His four grandchildren and the love and respect of his former students and colleagues helped him to recover from his deep sorrow. In 1961 the University conferred the degree of doctor on him; at the anniversary of 1963 a honorary degree was conferred on him and in 1965 received a special diploma for his 65 years professional activity.

His colleagues immortalized his memory by naming various plants after him: e.g. a *Chrysanthemum* sort, a blackberry—rapsberry hybrid named *Rubus*×*Mohácsyanus*, an interesting linden-tree named *Tilia Mohácsyana*, and a wild hybrid of dwarf morello and wild cherry found also in Austria named *Prunus (Cerasus)*× *Mohácsyana.*

In his last years he could see the enormous progress which was due to his life-work, too, and lived to see the University of Horticulture come into existence.

His biography entitled "Kert a parlagon" (Garden on the waste) was written by Endre Dobray dr. and the bibliography included compiled by Gusztáv Geday dr.

Z. E. KÁRPÁTI

Acta Agronomica Academiae Scientiarum Hungaricae, Tomus 21 (1—2), pp. 285—290 (1972)

RECENSIONES

LEN A KONOPÍ

8 1970

NEPERIODICKY VYDÁVÁ: VÝZKUMNÝ ÚSTAV
TECHNICKÝCH PLODIN A LUSKOVIN V ŠUMPERKU - TEMENICI
VE SPOLUPRÁCI S LNÁŘSKÝM PRŮMYSLEM, OBOROVÉ ŘEDITELSTVÍ, TRUTNOV

Len a konopi (Flax and hemp), 1970, 8 (103 pages)

Len a konopi a collection of papers written in Czech, is the joint publicaton of the Research Institute for Industrial Plants and Legumes, Sumperk-Temenice, and the Special Directorate of Linen Industry, Trutnov, on 103 pages, with many tables and pictures.

Among the fibre plants playing a very important role in Czechoslovakia the production, breeding and phytopathological problems of flax are dealt with in 7, and questions of hemp growing in 2 papers. Each paper is completed by German and English summaries and — except the first paper — literary references.

Trnka M. presents in 3 tables the results of national variety trials carried out with flax during 1968 in Czechoslovakia. Due to the unfavourable weather conditions the results were worse than in the previous years. "Vera" is the most efficient and wide-spread flax variety, certified ten years ago. After the three-year cycle of the state experiments had been completed the variety KO bred in Ceská Belá and the Soviet variety LD-147 were left out of the experiments. Investigations were continued with the variety 41 K/II bred in Ceská Belá and B-1 bred in Bystrica pri Martine.

Brejcha L. studies the effect of lodging on flax fibres in field experiments and by laboratory and technological analyses. The author points out that lodging affects the development of the whole plant including the internal structure of the stalk. He presents in four tables some characteristics of the flax varieties "Textilák", "Vera" and "Flachskopf" and the effect of lodging, and points out that lodging prior to flowering mainly damages the quality and quantity of fibres, while lodging after flowering has an unfavourable influence on seed production. The effect of lodging on the fibre cells is shown in three figures.

Láskos J. experimented with hemp in order to determine the effect of temperature on the germination power and germinative ability of the hemp varieties "Sumperská Alfa" and "Rastislavické" and of the monoecious line No. 65. The author gives the results in two tables which show that in practice the variety "Rastislavické" can only be sown when the temperature of the soil is over 10°C, while "Sumperská Alfa" and line No. 65 can already be sown at a soil temperature of 5°C.

Bernáth S. wanted to improve the technological quality of the fibres in the Slovakian hemp variety "Rastislavické" during a three-year agrotechnical experiment serie. By applying deep ploughing as well as organic and inorganic fertilizers to a clay soil poor in nutrients the author considerably increased the stalk yield of hemp and improved its quality. He supports his results with a figure and data included in five tables.

Ondrej M. makes a valuable contribution to the identification of the fungus *Kabatiella lini* (Laff.) Karakulin (= *Polyspora lini* Laff.) which causes the breaking and browning of flax stalks. The author presents in six figures the germinating conidia of the fungus, its damage to the cotyledons, its mycelium in the intercellular space, the development and separation of conidia, furthermore — for the purpose of comparison — the picture of the morphologically very similar *Pullularia pullulans.*

Hain J. and Procházka F. recommend a new breeding method for the variety maintenance of flax which requires more work from the breeder but reduces the costs. This method essentially consists of pure lines sown with capsules (capitula).

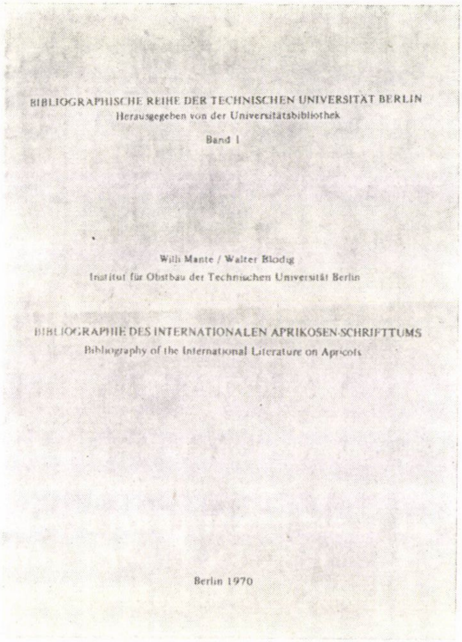
Pospisil B. in this second chapter of his previously published paper illustrates in 7 tables and 4 figures the way of obtaining the highest seed propagation coefficient of fibre flax. The optimum result was attained with 600—1000 germinating seeds per 1 m^2 sown at a row distance of 15 cm. Seed yield was the lowest with a row distance of 40 cm applied; while 7.5 and 30 cm gave about the same medium result. The fibre quality was the best with the smallest — 7.5 cm — row distance.

Procházka F. discusses a breeding method based on the pure line of fibre flax and called "Linie" method by him. This method is based on producing populations by crossing, increasing the frequency of mutants in the initial material, or the two processes combined. The described method is one of those used at the breeding station of Ceská Belá.

Zmeskal O. confirms the old theory, namely, that flax must not be sown to the same place year after year. The author carried on experiments from 1958 to 1968 with flax sown to the same place and overall fertilization applied. The germination percentage did not change until 1965, but in 1966 decreased by 50 per cent. The yield — on the other hand gradually decreased from year to year until 1966, when it almost failed. A microbiological analysis showed a mass propagation of pathogens in the soil, there are, however, other factors so far totally unknown that play a part in the self-incompatibility of flax.

The publication which discusses thoroughly several production and breeding problems of fibre plants provides many interesting data for the specialists. Finally it gives a necrology on Svitil Zdenek, the outstanding expert of flax, assistant director of the Research Institute for Industrial Plants and Legumes, and lists the research tasks solved by him.

B. FRIEDRICH



BIBLIOGRAPHISCHE REIHE DER TECHNISCHEN UNIVERSITAT BERLIN
Herausgegeben von der Universitätsbibliothek

Band I

Willi Mante / Walter Blodig
Institut für Obstbau der Technischen Universität Berlin

BIBLIOGRAPHIE DES INTERNATIONALEN APRIKOSEN-SCHRIFTTUMS
Bibliography of the International Literature on Apricots

Berlin 1970

W. MANTE, W. BLODIG: *Bibliographie des Internationalen Aprikosen-Schrifttums* (Bibliography of the International Literature on Apricots). Bibliographische Reihe der Technischen Universität Berlin, Berlin, 1970.

All kinds of scientific experimentation has to be preceded by tiresome and often very expensive literary research. However, as a result of a rapid development in almost all branches of science, for several reasons the special literature is becoming vaster and vaster. The cause is, on one hand, that only a small number of institutes possess all the important scientific reviews and technical journals of the world. On the other hand, in spite of the lengthy and tiresome literary research preceding the actual experimenting work, there is often a danger of important works closely related to the subject not becoming known to the researcher. That is often the case with papers published earlier, or not published at all (e.g. dissertations).

For a beginner in search of a subject indispensable information is given by a properly arranged documentation compiled possibly from a critical viewpoint too. At the same time those who have been working for some time can also get considerable help and further ideas from a bibliography compiled with thorough care.

A complex and intensive study of any problem requires an international co-ordination which is impossible without a detailed knowledge of the literature.

For the solution of the problems mentioned substantial help is given by Mante—Blodig's "Bibliography of the International Literature on Apricots", which provides, at the same time, a cross-section of the comprehensive literary documentation established by Willi Mante at the Fruit Growing Institute of the Technical University Berlin in 1957.

The excellent work can be divided into five parts: 1. Preface, 2. Introduction, 3. Papers grouped by special field, 4. Alphabetic list of authors, 5. Register of subject groups.

The book presents the titles of German and Roumanian papers in the original and — in certain cases — translated into English; titles of other foreign papers are translated into English. It is a pity that 39 of the 40 titles of the Hungarian papers contain bigger or smaller mistakes.

The bibliography contains a total of 2925 titles arranged in chronological order in two parts. The first part includes 2525 papers up to 1966, the second part 400 papers up to 1967/68. The chronological order of the papers at the same time gives a clear idea of the historical development of apricot production all over the world.

The publications are grouped according to the various special fields within which further smaller groups are formed in accordance with the identical subjects of the papers. The classification of the publications by special field was carried out taking their importance in consideration — not in every case unobjectionably. E.g. of the papers dealing with the water regime of the apricot only a few are found under the entry of "water regime", although this extremely important physiological process is dealt with by quite a lot of authors. The necessary information on the proper place of the major groups is given in the Contents.

The first special field covers papers of a morphological and anatomical nature. In comparison with the other groups this part is not significant. Except for a single study, the papers were published during the last ten years.

The chapter on metabolic physiology includes papers dealing with the nutrient supply and substances of various plant parts — with special regard to the amino acids and carotenes. Papers on the water regime of plants are also contained in this group.

A great many papers are written on the subject of development physiology. These papers deal with the effects of edaphic and climatic factors, the unfavourable ecological factors and the dormant period of the apricot. Within this group a separate sub-group contains studies on flowering, fruit production, bud differentiation, and chemicals with hormone-like activity as used for fruit thinning.

In the chapter on seed- and germination physiology the papers discuss the analysis of the seed and the vegetative development of seedlings.

Apricot breeding is dealt with in only a few papers. Beside the direct work of breeding, selection and crossing play an important role. Papers on frost resistance too are found in this group.

Beside the botanical grouping, in the chapter on propagation and plantation a considerable part is taken by the question of root propagation.

As to the question of variety and stock, a number of papers unanimously suggest that each variety should be planted to a definite place.

The large number of papers suggests that apricot planting is dealt with in many countries. Unfortunately, no uniform picture can be obtained owing to the differences in methodology.

Numerous papers deal with plantation technics, and within this with the proper choice of soil, crown shapes, pruning techniques, plant and soil cultivation, weed control. In this group are included the papers discussing irrigation and fertilization too.

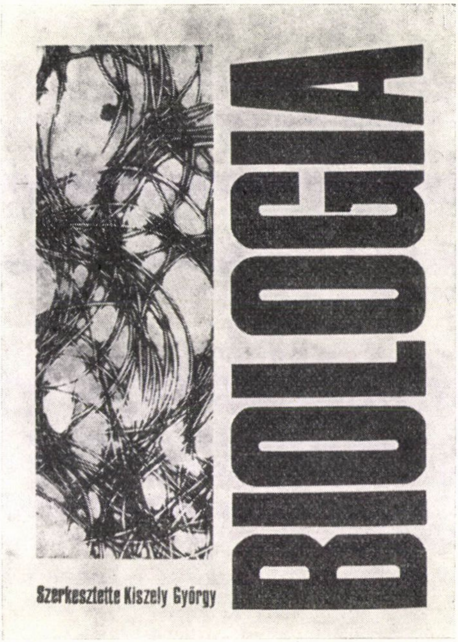
As to the content of the papers, one of the most important parts is related to plant protection. Many papers deal with the diseases of the plant, first of all of a virus origin, but there are quite a lot discussing animal pests and the protection against them too.

Papers related to fruit harvesting and storage are much less in number. Studies of an economic nature as well as those concerning processing are of a similarly low number.

The alphabetic list of authors found at the end of the bibliography, as well as the register of subject groups make the bibliography very easy to handle, and give much help in literary research.

By their excellent work "Bibliography of International Literature on Apricots" compiled with thorough care Willi Mante and Walter Blodig, co-workers of the Library of the Technical University, Berlin, provide great assistance to many a researcher, and have opened up new vistas, which rightly arouses international interest and acknowledgement.

Gy. Borka



Gy. Kiszely, T. Ács, Gy. Csaba, G. Szabó: *Biologia* (Biology). Medicina Könyvkiadó, Budapest, 1970.

General biology is the basis of the special sciences of applied biology. Modern biological text-books of a cytological, genetical and biochemical approach have — especially today — an important role, since practical experts (doctors, veterinarians, agronomists, chemists etc.) can only find their ways in the great mass of special information if they have an adequate level of biological foundation. The text-book discussed here promotes biological education at medical universities, but gives useful information to all experts who are interested in biology.

In Hungary it was Tivadar Huzella who laid the foundation of medical biology, then text-books on medical biology were published twice (in 1956 and 1966) under the editorship of Imre Törő. The recently published Biology not only summarizes the great results attained so far, but also discusses the basic relations of biology with an excellent conciseness.

The book consists of 542 pages, with numerous original and borrowed figures and clear tables, as well as excellent photos. It is divided into the following nine main chapters:

1. *Biology and the medical way of thinking.*

2. *Concepts of evolution, complexity and organization in nature.*

3. *Abiotic evolution and organization of material.* Atom and its organization; Relative frequency of occurrence of atoms in inanimate nature and living organisms; Biogen elements, biogen compounds; Specificity of biogen compounds; Possibility of abiogen synthesis of carbon compounds; Rudiments of colloidy and biophysics; Molecular functions.

4. *Biological organization.* Origin of life; Biological organization and individuality of life; Concepts of protoplasm and cell; Unity of structure and function. The concept of molecular biology; Structure and function of the cell (microscopic and submicroscopic functional morphology of the cell; cell functions and their control; multiplication of cells; general genetical concepts; Cytogenetics); Molecular genetics; Cell differentiation (phenogenetics at the level of cells); Levels of biological organization.

5. *Organism and its environment.* Ecological elements; Relations between organism and its environment; Homoiostasis of the organism, and the environment; Guidance and control in the organism.

6. *Reproduction and sexuality.* Concepts of reproduction and sexuality. Vegetative and sexual reproduction; Seed-phase change and alternation of generation; Definition and structure of sex.

7. *Basic phenomena of ontogenesis.* Biological concept of ontogenesis; Development and growth; The genetics of ontogeny; Morphogenetic processes; Regeneration; Senescence, Life span, Death; Biological relationship between mother and embryo; Pathological development, Biological bases of teratogenesis.

8. *The elements of human genetics.* Relationships between gene and phene; Test methods of human genetics (statistical–mathematical method, twin research, genea-

logical analysis, cytogenetical method, biochemical method); Fight against hereditary diseases; Genetical, environmental and medical implications of the habitus; The concept of anthropology.

9. *Biological evolution.* Evidences of biological evolution; Genetical problems of evolution; Ontogenetic relations of evolution; The birth of new species; The origin and future of man.

Although the book has not been written for agriculturists, it still may be very useful for all practical and theoretical workers engaged in agriculture (veterinarians, live stock breeders, toxicologists, agricultural biology teachers etc.), since it gives answers to important questions, such as the relationship between damages done by chemicals and teratogenesis, or lifetime as closely related to nutrition, the biological relations of senescence etc. The great number of analogies that experts with botanical views may find while studying the book — may be of similar interest.

Through the concise and clear treatment of the most general and fundamental relations of biology a valuable monography has been added to the Hungarian general biological literature.

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1828

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AUGUST 1971 VOLUME 27 NO. 1

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РЕЗЮМЕ

СОДЕРЖАНИЕ ФОСФОРА, ЛИПИДА И ФОСФОЛИПИДА В МИОФИБРИЛЛЯРНЫХ БЕЛКАХ

Содержание липида и фосфора в миофибрилле и актине

Ш. ФАЗЕКАШ, В. СЕКЕШИ-ХЕРМАН, Л. ВОДНЯНСКИ

В работе изучали содержание фосфора, суммы липидов и фосфолипидов в миофибрилле и актине. В опыте установлено, что миофибрилл и актин обладают значительным содержанием липида, поэтому вес, высчитанный на основе измерения содержания белка, отстает от веса, измеренного путем гравиметрии. Часть биурет-негативного привеса устраняется в ходе очистки миофибрилла, но большую часть компонентов, содержащих фосфор, нельзя устранить описанными методами. Привес, обнаруженный в актине, частично устраняется при очистке путем де- и реполимеризации и ультрацентрифугированием, а другая часть — обработкой Dowex 50, экстрагированием в смеси Chl : MeOH или гель-фильтрацией. Из актина, полученного путем гель-фильтрации, можно изолировать около 10 процентов липида. Ожидается, что молекулярный вес 46 000 г/мол, известный до сих пор, окажется немного меньшим. Изменение в размере молекулы, уже не обладающей способностью к полимеризации актина, полученного при обработке Dowex 50, можно обнаружить при гель-фильтрации на колонке Seph. G 200; пик передвинется слегка вправо по сравнению с актином, обладающим полимеризационной способностью. Липидные компоненты миофибрилла и актина на воздухе подвергаются автоокислению так, что их цвет и положение на пластинке ТЛЦ изменяются соответственно степени окисления; как и ожидалось, в миофибрилле можно было найти больше, а в актине меньше компонентов липида. При очищении актина уменьшение компонентов липида приводит к увеличению полимеризационной способности, но устранение липида, вызывающее нарушение структуры, прекращает полимеризационную способность актина и приводит к гетерогенности белковой части.

АНАТОМИЯ ВЕГЕТАТИВНЫХ ОРГАНОВ, НАКАПЛИВАЮЩИХ ЗАПАСНЫЕ ВЕЩЕСТВА, II. СТЕБЛИ

Г. Ш. ПАЛИВАЛ, А. К. КАВАТХЕКАР

Изучалась анатомия вегетативных органов, накапливающих запасные вещества у растений *Amorphophallus campanulatus* и *Colocasia esculenta*. Оба имеют толстый перидерм. В массивной диффузной паренхиме имеется немного разбросанных сосудистых пучков, большое количество крахмальных зерен и многочисленные изолированные латексные клетки. Рассматривается их структуральная модификация в связи с выполняемой ими функцией.

ОСОБЕННОСТИ МЕМБРАНЫ У ПОРЫ ПЫЛЬЦЕВОГО ЗЕРНА SOLANUM DULCAMARA L: I.

Распад мембраны

Б. БАРНАБАШ, ДЬ. ПАЛ

На основании ультраструктурных исследований предполагается, что на месте поры пыльцевых зерен цитоплазматические трабекулы, пронизывающие интину, а также и их выпячивающиеся из интины части ограничиваются плазматической мембраной. Эта гипотеза подтверждается нашими наблюдениями, выполненными на пыльцевых зернах *Solanum dulcamara L.*, по которым в случае, когда кальций, обеспечивающий стабильность этой мембраны, оттягивается из структуры мембраны с помощью связывающих кальций соединений, стабильность мембраны прекращается. В этом случае мембрана распадается, а цитоплазма вегетативной клетки вытекает через пору и растекается. Наряду с доказательством существования мембранной структуры у поры, наше наблюдение имеет и практическое значение. Во время цветения, а также выбрасывания пыльцевых зерен из пыльников нельзя применять гербициды, которые связывают кальций, обеспечивающий стабильность плазматической мембраны, так как они могут проникнуть в плазматическую мембрану интины, расположенной под порой. То есть, то пыльцевое зерно, у которого плазматическая мембрана распадается у поры и цитоплазма вегетативной клетки вытекает через пору и растекается, не способно ни к прорастанию, ни к оплодотворению.

ВЛИЯНИЕ ОБРАБОТКИ ССС НА СЕЯНЦЫ РАЗНЫХ КОСТОЧКОВЫХ ПОРОД

Д. ШУРАНИ

Проводилась обработка сеянцев дикой черешни, а также антипки препаратом ССС при концентрациях 0, 500, 1000, 2000 и 4000 ppm. Перманентное задерживающее влияние роста наблюдалось только у сортов с большой силой роста (дикая черешня и миробалан). Эффективность ССС коррелировала с силой роста. Положительная корреляция наблюдалась также между задержкой роста стебля и увеличением содержания сырого белка.

МЕТОД ВЫБОРА СУЩЕСТВЕННЫХ ПРИЗНАКОВ

Ш. ЙОЖА

Изучая несколько признаков, более или менее изменяющихся в зависимости друг от друга, оказалось, что некоторые из них вероятно можно выпустить без потери значительной информации. Редукция числа признаков в этом смысле является очень желательной со многих точек зрения. Несмотря на это в специальной литературе нет приемлемого метода для выбора несущественных переменных. В данной работе показаны основные принципы одного возможного метода.

ДЕЙСТВИЕ ЦИТОСТАТИЧЕСКОГО ДИБРОМОМАННИТОЛА НА СИНТЕЗ ПРОТЕИНА В МИЦЕЛИИ BOTRYTIS CINEREA И SCLEROTINIA TRIFOLIORUM

Л. ДЬ. САБО, Л. ХОЛЛИ, Л. ХОРВАТ, Б. И. ПОЖАР

На культуре мицелия двух изучаемых микрогрибов — фитопатогенов: **Botrytis cinerea Pers.** (деутеромицеты) и **Sclerotinia trifoliorum Erikss.** (аскомицеты) авторы доказывают влияние цитостатического дибромоманнитола на увеличение уровня белкового азота по отношению к содержанию сухого вещества путем измерения инкорпорации радиоактивного углерода, маркирующего глицин в протеинах мицелия. Характерно, что дибромоманнитол увеличивал отношение белкового азота к содержанию сухого вещества при всех примененных концентрациях, тогда как в других опытах угнетал синтез дезоксирибонуклеиновой кислоты. Более чем 20 процентное изменение в пропорции протеиновой фракции является важным и с теоретической точки зрения.

ПРИМЕНЕНИЕ КВАНТИТАТИВНОГО АНАТОМИЧЕСКОГО МЕТОДА ДЛЯ СРАВНЕНИЯ СОЛОМЫ НЕКОТОРЫХ СОРТОВ ПШЕНИЦЫ

Й. ШТИБЕР

Автор исследовал сорта венгерской пшеницы Банкути 1201 и итальянской Сан-Пасторе, применив метод сравнительной квантитативной анатомии. Было определено, что у Сан-Пасторе длина волокон больше. Автор, совместно с сотрудником, вычислив разработанный ими фактор устойчивости (Т-фактор) показали, что способность к полеганию у пшеницы Сан-Пасторе ниже. Все эти результаты ввиду небольшого количества исследованных экземпляров (по 2 экземпляра от сорта), в первую очередь характеризуют отдельные индивидуумы; для обобщения необходимо провести больше исследований. В данной статье автор, в первую очередь, хотел показать метод. Анатомический метод, повидимому, может быть пригодным для числового выражения склонности пшениц к полеганию и для их группировки. Автор приводит различные определения и заключения по поводу закономерности изменения длины волокна и объема клеточной стенки.

ОБРАЗОБАНИЕ ПЫЛЬЦЕВЫХ ТРУБОК У ГРУШИ

Й. НИЕКИ

В 1968 и 1969 гг. у сортов груши «Clapp kedveltje» и «Vilmos» были изучены: зависимость между образованием пыльцевых трубок и концентрацией сахарозы, продолжительность образования пыльцевых трубок и влияние температуры. Оба сорта образовали максимальный процент трубок при концентрации сахарозы в 15 процентов. Между процентом образовавшихся пыльцевых трубок и продолжительностью проростания пыльцы обнаружена линейная корреляция. Образование пыльцевых трубок достигало максимума за 120 и 160 минут, соответственно. Зависимость между ратой образования пыльцевых трубок и температурой можно иллюстрировать оптимальной кривой. Оптимальная температура для образования пыльцевых трубок была 23°C для сорта «Clapp kedveltje» и 25°C для сорта «Vilmos».

НОВЫЙ ИСКУССТВЕННЫЙ ГИБРИД МЕЖДУ РОДАМИ FESTUCA И LOLIUM (FESTUCA PRATENSIS HUDS. X LOLIUM TEMULENTUM L.)

Л. ХЕСКИ

В 1969 и 1970 гг. были скрещены виды из двух родов в различных комбинациях. После искусственной кастрации, изоляции и опыления цветков часть гибридных эмбрионов воспитывалась in vitro, гибриды получены в нескольких вариантах, один из которых оказался особенно ценным. Эта межродовая форма была получена от скрещивания ***F. pratensis*** (2×) и *L. temulentum* (2×). В статье представлены наблюдения, связанные с гибридами.

ВАЖНОСТЬ ВЗАИМОДЕЙСТВИЯ В ПОВЫШЕНИИ СОДЕРЖАНИЯ ПРОТЕИНА У МУТАНТНЫХ ПОПУЛЯЦИЙ

Я. СИРТЕШ

Для того чтобы индуцировать генотипическую изменчивость по содержанию протеина, озимый ячмень сорта «Хорпачи кетшорош» с низким содержанием протеина был подвергнут обработке ЭМС. Содержание протеина изучалось на растениях M_2 с фертильностью колоса 90—100% и их линиях M_3. Среднее содержание протеина в популяции M_2 и M_3 было на 29% выше, чем в контроле с низким содержанием протеина (11,2%). В генетически гетерозиготной популяции мутантов варианса взаимодействия между генотипом и годом была значительно высокой по сравнению с вариансами генотипа и ошибки. Поэтому, в этой популяции от селекции мог ожидаться только средний прогресс. Затем мутантная популяция была разделена на популяции по взаимодействию между генотипом и годом (низкое и исходное). Среднее содержание протеина обеих популяций оставалось совершенно одинаковым, но предсказанный генетический прогресс улучшился, в очень высокой степени у популяции с низким взаимодействием. Эффективная селекция, таким образом, требует не только благоприятной средней линии, но также и низкого взаимодействия между генотипом и годом.

НОВЫЙ МЕТОД НАБЛЮДЕНИЯ ЗА РОЕМ ОЛЁНКИ (EPICOMETIS HIRTA PODA)

Ф. КОЗАР

В течение 1970—1971 годов с целью изучения динамики роения личинок. *E.* hirta выработан новый метод прогноза с помощью цветной ловушки, состоящей из сосуда, наполненного подкрашенной в синий цвет водой. При испытании синий цвет, похожый на окраску василька, удачно приманивал вредителей. Белый, желтый, зеленый и красный цвета не оказали приманного влияния. Сосуд синего цвета способствовал сбору и разных видов Hymenoptera, встречающихся во фруктовом саду. На улов в значительной мере влияли микроэкологические условия, поэтому необходимо применять больше ловушек для изучения распространения E. hirta в данной культуре.

ВЛИЯНИЕ ИНТЕНСИВНОСТИ СВЕТА НА ПРОДУКТИВНОСТЬ ТОМАТОВ (LYCOPERSICUM ESCULENTUM MILL.)

С. Р. БАРООВА, К. САС, И. ХОРВАТ

У двух сортов томата, выращенных в поле при двух вариантах интенсивности освещения, изучалось содержание хролофилла, вес сухого вещества, накопление углеводородов и азота. При высокой интенсивности освещения увеличились вес сухого вещества, содержание хлорофилла и углеводородов, но уменьшилось содержание азота в растениях. Сорта «Кечкеметский консервный» и «Кечкеметский карликовый» показали сравнительно лучшую продуктивность при самой низкой интенсивности освещения в условиях затенения.

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The Editors

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PHOSPHORUS-, LIPID- AND PHOSPHOLIPID CONTENT OF MYOFIBRILLAR PROTEINS. I

LIPID- AND PHOSPHORUS CONTENT OF MYOFIBRIL AND ACTIN

By

S. Fazekas, V. Székessy-Hermann, L. Vodnyánszky

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The paper deals with the phosphorus-, total lipid- and phospholipid content of myofibril and actin. In the authors' experiments myofibril and actin were found to contain a considerable amount of lipid, so the weight calculated on the basis of the measured protein content was lower than the gravimetric value. A part of the biuret negative weight surplus is removed when the myofibril is purified, but most of the components containing phosphorus cannot be removed by the methods described. The surplus weight found in actin is removed during the purification partly by de- and repolymerization and ultracentrifuging, partly by Dowex 50 treatment, extraction with a mixture of Chl : MeOH, or gel filtration. From the actin obtained by gel filtration some 10 percent lipid can be isolated. Thus, the molecular weight at present thought to be 46000 g/mol is probably somewhat lower. In the molecular weight of actin obtained by Dowex 50 treatment and no longer possessing a polymerizing ability this change can be observed during gel filtration on a Seph. G. 200 column, if the peak shifts slightly to the right compared to that of the polymerizing actin. The lipied components of myofibril and actin autooxidize in the open air, thus their colour as well as their position shown on the TLC plate change according to the degree of oxidation; myofibril contains more, and actin less lipid components — as was to be expected. During the purification of actin the decrease of lipid components results in an increased polymerization ability, but the removal of lipids, which causes the disintegration of the structure, puts an end to the polymerization ability of actin and leads to the heterogeneity of the protein fraction.

Introduction

A vast literature deals with the phosphorus metabolism of the muscle, and there are abundant data on its phosphorus and lipid content too, though they are — as yet — not sufficiently detailed and differentiated. It was only in the last ten or fifteen years that the determination of the categories and amounts of muscle lipids became possible by applying more refined methods of differentiation — such as paper-, layer- and column chromatography, solvent complexes and specific reactions. Though earlier the muscles had been thought to contain very little — if any — lipids, and "fatty muscle" to be a degenerative phenomenon, the idea that muscles also contain a considerable amount of lipid recurred more and more definitely, however, while the earlier statement was considered right it was found that "The intracellular muscle lipids are not used as a net source of fuel for the increased energy metabolism of the contracting muscle. It seems paradoxical that the intracellular muscle lipids should serve as an energy reservoir . . ." (Masaro 1967).

Many authors took part in detecting the muscle lipids and their components and developing the suitable methods (MARINETTI *et al.* 1957, GRAY—MACFARLANE 1961, MASARO 1967, MASARO *et al.* 1964, FRÖBERG 1967, KOSTRIKINA—EPSTEIN 1965). The fatty acid composition of muscle lipids has been determined in various muscles (CRAWFORD *et al.* 1970, DVORÁKOVÁ—BASS 1970a, DVORÁKOVÁ—BASS 1970b, OWENS—ANGELINI 1970, SIMON—ROUSER 1969). The findings suggest that the lipids and phospholipids may have an important role in some components of the muscle, such as: mitochondrium, microsoma, sarcoplasmatic reticulum, nucleus and cell membrane.

The presence of lipids in the myofibril was first suggested by SZENT-GYÖRGYI (1951), then GUBA (1954) and GARAMVÖLGYI—GUBA (1967) published data confirming the lipid content of myofibril. GUBA (1967) found a 5 per cent lipid content in the fibrilline itself (a myofibrillar protein isolated by him).

In contrast to the vast number of literary data concerning the phosphorus metabolism of the muscle, hardly any data are available on the phosphorus metabolism and absolute phosphorus content of the myofibril. Data on the absolute phosphorus content of myofibril contain contradictory results, since the authors start from different aspects and set different aims when trying to decide the question. The phosphorus content of myofibrillar proteins too is discussed only in two publications (MARINETTI *et al.* 1957, LYNN 1965), while changes in the phosphorus content have been dealt with by many authors.

For analytical reasons the earlier data on the total lipid content of muscle were not reliable. While earlier authors reported a 0.2—2 per cent lipid content, recent determinations give account of 6—8 per cent (MARINETTI *et al.* 1957, SERAYDARIAN—MOMMAERTS 1965). The difficulties of determination are further increased by problems of isolation and analysis when the lipid content is examined in some subcellular units of the muscle.

The only information on the lipid content of myofibril was given by GRAY—MACFARLANE (1961) who found 20 per cent lipid in the muscle myofibril. The lipid content of myofibrillar proteins has not sufficiently been studied; sporadical data of varying reliability either deny the low amount of lipid proved to occur in the structure proteins or consider it a contamination. Observations made in the last several years at our Institute in the course of protein isolation proved the presence of lipids. From the myofibrillar proteins — when dialyzed against an adequate solution — low-molecular-weight components of non-protein nature get into the dialyzing solution, which in the ultraviolet spectral range have a characterless flat spectre showing no maximum. Low-molecular-weight components isolable from actin during purification were discussed earlier (FAZEKAS *et al.* 1967a).

The present paper gives further data on the absolute phosphorus content of myofibril and myofibrillar proteins, and on the amount and composition of lipid components.

Material and Method

A purified myofibril suspension was prepared from the back- and leg muscles of four months old rabbits by the method of PERRY—CORSI (1958).

Actin preparations. The polymerizing tropomyosin free G-actin was obtained from aceton powder through extraction with CO_2-free icy distilled water (DRABIKOWSKI—GERGELY 1962, MARTONOSI 1962), polymerization with 0.3—6 mM $MgCl_2$, and dyalization in the presence of 2.5×10^{-4} M 2-mercaptoethanol after MARTONOSI (1962). "Free" nucleotid- and phosphorus contents of protein solutions were removed by Dowex II-X-8 treatment (DRABIKOWSKI—PISAREK 1964).

Actin containing minimum quantities of phosphorus and lipid was obtained after polymerization and depolymerization by Dowex II-X-8 treatment (ASAKURA 1961a) without gel filtration. Dowex 50×4 treatment was applied to remove contamination from the surface of the bivalent cations and other molecules as suggested by DRABIKOWSKI—PISAREK (1964). The pH-value of the Dowex 50 suspension was fixed at 7.2 immediately before use, and cca. 1—2 mg suspension was added to each ml protein (which contains 3—6 mg/ml protein), then, immediately after having been mixed, centrifuged cold (5 minute, 1 000g). By repeated treatments the lipid content was reduced to 7—10 percent, and the phosphorus content to 3.7—4 g atom/mol protein (60 000 g)*, when the polymerizing ability still remained unchanged. Then the ultra-violet absorption value of 1 mg actin corresponded to an E_{290} value of 1.095 in case of pH 13 (if the protein content was determined by the Kjeldahl method). Spectral characteristics and ΔE spectra are identical with those described earlier (FAZEKAS *et al.* 1967b).

Finally, the actin was purified by gel filtration on a Sephadex G 200 column (on standard column 2.3×52 cm). The Sephadex column was equilibrated with a mixture of 2.5×10^{-4} M ATP and 2.5×10^{-4} M 2-mercapto-ethanol and eluated with the same solution. (From a protein solution of 5—6 mg/ml concentration maximum 12 ml can be placed on the column.) The elution maximum of actin was obtained at an elution constant (kD-value) of 0.82. Elution constants (kD) were calculated by GELLOTTE (1960).

The homogeneous actin* was isolated with an extracting solution containing ATP according to the method of REES—YOUNG (1967). The extracting solution contained 5×10^{-4} M ATP, 5×10^{-4} M 2-mercapto-ethanol and 2×10^{-4} M Ca ion. The actin was gel filtrated from the standardized column with an eluating solution used by REES—YOUNG (1967). Polymerizing actin fractions were obtained at peak III, at a kD-value of 0.81—0.83.

From a myofibrillar suspension the actin was obtained from the myofibrillar residue treated four times with the myosin extracting solution of HASSELBACH—SCHNEIDER (1951) by the method of MARUYAMA (1966), and with an extracting solution containing ATP after HAMA *et al.* (1965) respectively, then purified. In both cases large amounts of myofibrillar suspensions were used, since the above methods yield — at a very low degree of efficiency — small quantities of "pure" actin.

The quantitative determination of the initial material was performed by the concentration of an aliquot solution and drying at 105°; then, after the isolation and dialyzation of the individual fractions the procedure was the same as before. The lipid-free protein isolated from the fractions were repeatedly dialyzed, then dried in a vacuum thermostat over cc. H_2SO_4, and gravimetrically measured. The protein content was determined — from the nitrogen content measured by the Kjeldahl method — either by GOA's microbiuret method (1963), or spectrophotometrically, from the extinction coefficient measured at 280 nm (pH 7) and 290 nm (pH 13) respectively.

Lipid extraction. From purified myofibril, and actin preparations respectively, the lipid was extracted with FOLCH's solution (1952) modified by FOLCH *et al.* (1957). The first extraction was made with a 10 vol. solution during one and a half days, the second with 8 vol. in one and a half days, while the third extraction was made with a 5 vol. solution for one day. Thus the extraction takes four or five days. The extracts usually contain an insignificant amount of biuret positive substance called crude lipid. The extracts were distilled dry in a rotary evaporator, resolved in'a 20 vol. solution of chloroform: methanol (2 : 1 v/v), centrifuged, and then the biuret positive substance precipitated. The procedure involves a substantial loss of material. A considerable proportion of unsaturated lipids was removed by washing with water, therefore the above method had to be completed by other methods and modifications. They will be discussed together with the respective preparations.

Lipid extraction from dry gel. In some cases the dyalized fractions, or protein prepara-

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* The molecular weight of actins obtained without gel filtration was determined in 60 000 g according to an earlier calculated value.

tions were treated — after perfect concentration in a vacuum thermostat, or drying at 105 °C — with an extracting solution of chloroform : methanol : water (65 : 33 : 2, v/v/v). From the dry gel usually protein-free, biuret negative lipids are obtained, though of a much lower quantity than when extracted from wet preparations.

Nucleotide determination in actin preparations was carried out by the method of ASAKURA (1961b). First the free nucleotide was removed from the actin solutions, then the actin-bound nucleotide was released with perchloric acid. The perchloric acid was removed by KOH precipitation from an icy solution and centrifuging. Finally, the nucleotide was determined by spectrophotometry. The purity of the nucleotide was controlled in one or two cases by separation on a Dowex II-X-8 (form Cl) column.

The total phosphorus content of the samples was determined after destruction in a mixture of cc. $H_2SO_4 : HNO_3$ (1 : 2, v/v) according to FISKE—SUBBAROV (1925), but the final reduction (to a volume of 1 ml/10 ml) was achieved by ascorbic acid treatment after LOWRY *et al.* (1954).

The cholin content was determined by the method of APPLETON *et al.* (1953). The aliquot was taken from the hydrolysate and the HCl concentration reduced through dilution to 2 N; then the organic acids were extracted first with 0.4 ml n-butanol, then with 3×2 ml isobutanol per ml, and the cholin in the aqueous residue was determined.

The lipids were separated by thin-layer chromatography, after WAGNER *et al.* (1961); or were developed either with a mixture of chloroform : methanol : 25% NH_3 (17 : 7 : 1, v/v/v) according to CUZNER—DAVISON (1966); or first the phospholipids were developed with SKIPSKI *et al.*'s (1963) system by a stepwise development method, and the neutral lipids led on. The chromatograms were coloured with iodine vapour (BRANTE 1949), or 0.01 per cent rhodamine G and detected in ultra-violet light, while in other cases they became visible after having been charred with 40 per cent H_2SO_4 and heated to 250 °C.

Results

Phosphorus- and nucleotide content of myofibril. Myofibril dried at 105 °C was found to contain an average amount of 660 μg/g (21.5 micromol) phosphorus. The nucleotide content as well as the phosphorus content extracted with perchloric acid were determined. The results are shown in Table 1.

Table 1

Phosphorus and nucleotide content of myofibrillar suspension in 1 g dry matter

Treatment	P-content microatom/g	Nucleotide content micromol/g	P-content after perchloric acid extraction microatom/g	P-content after 5 days dialysis microatom/g
1	20.7	1.45	16.2	16.1
2	21.8	1.8	17.1	18.0
3	22.0	1.55	18.0	17.7
Mean	21.5	1.6	17.1	17.1

The experimental data show that no more phosphorus can be removed by dialysis from the residue extracted with perchloric acid, and a considerable amount (550 μg = 16.2 —18.0 microatom/g) of phosphorus is left behind after drying at 105 °C. Only one quarter of one fifth of the phosphorus content is nucleotide.*

* The nucleotide content of myofibril was earlier measured by PERRY (1952) who determined an acid-labile phosphate by a seven minute hydrolysis and found it to be 90—140 g P/g. When converted into ATP this amount agrees with our results.

Total lipid-, phospholipid- and phosphorus content of myofibril. Extractions were made from myofibril with Folch's solution on three successive occasions, and the extractable lipid content computed after concentrating and redissolving the lipids, in a 20 vol. mixture of chloroform:methanol (2 : 1, v/v). 1200 mg total lipids were separated on silica gel column and only 148 mg (12 per cent) of them were found to contain phosphorus.

Table 2

Lipid content in purified myofibrillar suspension. Extraction: with modified FOLCH *(1957) solution*

Treatment	Percentage of extracted lipid			Total lipid %
	I.	II.	III.	
1	14.4	3.2	0.017	17.6
2	17.7	2.3	0.01	20.0
3	19.0	0.8	—	19.8
4	6.0	0.2	—	6.2
5	9.11	2—0	0.01	11.1

Table 3

Total lipid and phospholipid content in purified myofibrillar suspension after repeated washing and purification by* FOLCH'S *method***

Myofibril	Total lipid mg	Phospholipid	
		mg	%
1	1240	148	12.2
2	1567	133	8.5
3	1150	92	8.0
4	1036	84	8.2

* Related to total lipid.

** Myofibrillar lipids were separated from polar lipids on a silica gel column after PRIES *et al.* (1966).

The rest consisted mostly of fatty acid, di- and tri-glyceride (and some unidentified substance; see later). Cholesterin could be detected, if at all, only in negligible quantities in one or two cases. Table 2 contains the lipid-, while Table 3 the total lipid- and phospholipid content of the myofibril suspension.

In our experiments the lipid content of the myofibril suspension was somewhat lower than that indicated by the literature. However, repeated washing in weak borate buffer considerably decreases not only the protein- but also the lipid content of myofibril. While after washing twice, dialyzing in 0.01 TRIS-HCl (pH 7.8) buffer and centrifuging, about 30 percent of the total proteins is removed, in the case of washing seven or eight times in borate buffer, substance of only 0.1—0.6 E_{280} value is removed, even when examining the supernatant spectrophtometrically, and after dialyzing against TRIS-buffer for a further two days and centrifuging, only two or three percent of the total proteins can be found in the supernatant.

Nucleotide-, phosphorus- and lipid content of actin. Actin extracted for 20 minutes from acetone-dried powder and purified by a single polymerization still contains a high and varying amount of phosphorus (9—13 μg atom P) and 18—20 percent lipid related to the earlier determined 60 000 g molecular weight.

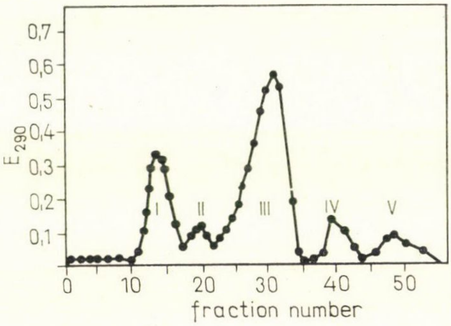

Fig. 1. Gel filtration of actin on Sephadex G. 200 column by REES-YOUNG (1967) method

The phosphorus content of actin treated with Dowex II and 50 ion-exchangers is lower, and by a repeated careful Dowex 50-treatment the total phosphorus content can be reduced to a minimum amount of 3,6—4.5 g atom and 5—10 percent extractable lipid content, when actin still retains its polymerizing ability. The molecular weight of actin isolated and gel filtrated according to REES—YOUNG (1967) is 46 000, and it is electrophoretically homogeneous (Fig. 1). In our examinations perchloric protein when determined by the method of ASAKURA (1961a) after precipitation contained 0.9—1.0 mol nucleotide, an average of 10 percent lipid and 0.5—1.0 mol phospholipid. In most cases some phosphorus is still left behind in the extracted protein, which after being reduced to ashes shows a phosphorus content of about 0.5—1.0 g atom.

Actin obtained from a myofibrillar suspension contained 6.9 g atom total phosphorus/60 000 g protein and 12—20 percent lipid. In this actin no nucleotide was found, it could not be detected after treatment with perchloric acid. The spectra is characterless, the small amount of nucleotide may remain covered, nevertheless it is this actin which is bound to the myosin and increases its ATP-ase activity.

The myofibrillar residue left behind after the extraction of actin contains a significant amount of phosphorus (438—466 μg = 14.1—15 micromol/g) even after repeated washing and a protracted dialysis.

Rees-Young's actin just like Straub, Martonosi's (Straub 1943, Martonosi 1962) actin can be transformed by Dowex 50-treatment into forms containing lower amounts of total phosphorus and lipid, and become similar to Dowex 50-treated polymerizing-, and Dowex 50-overtreated, no longer polymerizing actins respectively (Fig. 2).

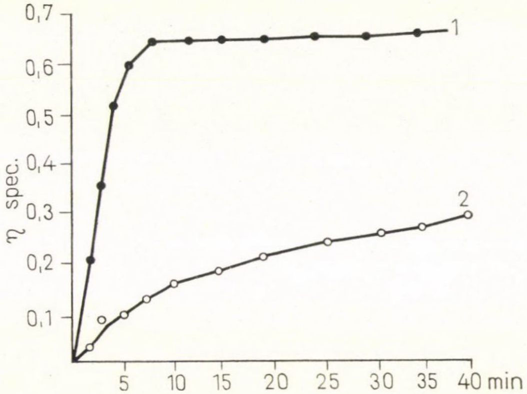

Fig. 2. Polymerization of fraction III of gel filtrated actin. 1. Actin containing about 1.1 M lipid phosphorus; 2. actin containing about 0.5 M lipid phosphorus. Flow time of the viscometer (Ostwald-type) was 29.1 sec per 2 ml at 24 °C

Actins with reduced polymerizing ability eluate during gel filtration the same or slightly higher (0.82—0.90) kD-values than the polymerizing actin of Rees—Young (1967).

All actins discussed so far when lyophilized and concentrated in vacuum thermostate become a light brown, slightly glassy gel no longer soluble only swelling in aqueous solutions. After several days swelling in distilled water 0.6—1.1 mg/ml protein is redissolved in the supernatant.

When dialyzed against distilled water all actins release low-molecular-weight acidic substances which enter the dialyzing water. After concentration the low-molecular-weight products show a low optical density and flat characterless spectra. They usually contain a biuret negative (or some biuret positive) substance. Concentrated low-molecular-weight substances show 3—4.5 pH-values. Their ultra-violet spectra are similar to those of the lipids of myosin, their ultraviolet absorption increases while they are stored (Fig. 3).

After standing for a long time the isolated lipids change their colour to yellow or brown.

Under the influence of a protracted dialysis the spectrophotometrically measured E_{280} value of the protein fraction increases in spite of the removal of low molecular-weight substances, and a 5—10 per cent surplus appears. The

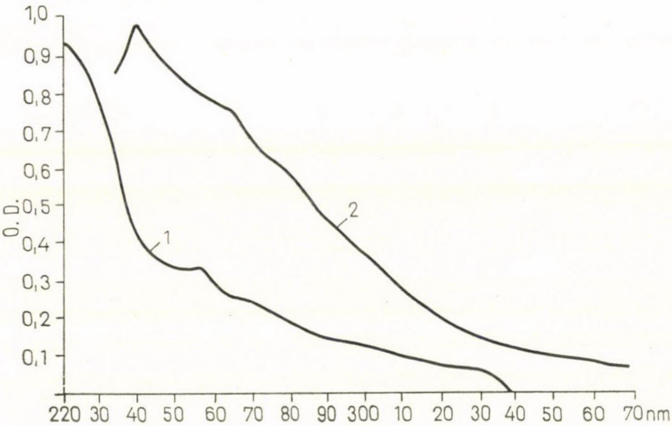

Fig. 3. Spectra of dialyzable lipid fractions from actin. 1. Immediately after isolation; 2. after 48 hours storage at 4 °C

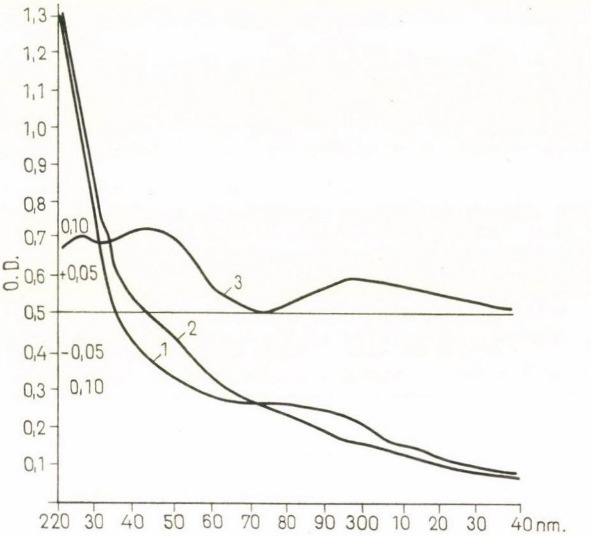

Fig. 4. Spectra and difference extinction spectra (3 ΔE) of aggregated F-actin, at pH 7 (1) and pH 13 (2)

dialysed protein solutions become heterogeneous and after standing for a while, sooner or later aggregate.

Inhomogeneity can be induced by Dowex 50 treatment. The spectra of aggregated actins change and become highly different from those of the normal actins (Fig. 4).

Substances of non-protein nature present in actin preparations can be demonstrated even by paper-chromatography. Whatman N^0 1 paper is washed in a mixture of ethanol: water (2 : 1, v/v), then placed in the solvent mixture without samples in order to remove contaminations, the — with the sample placed and developed — a chromatogram shown by Fig. 5 is obtained.

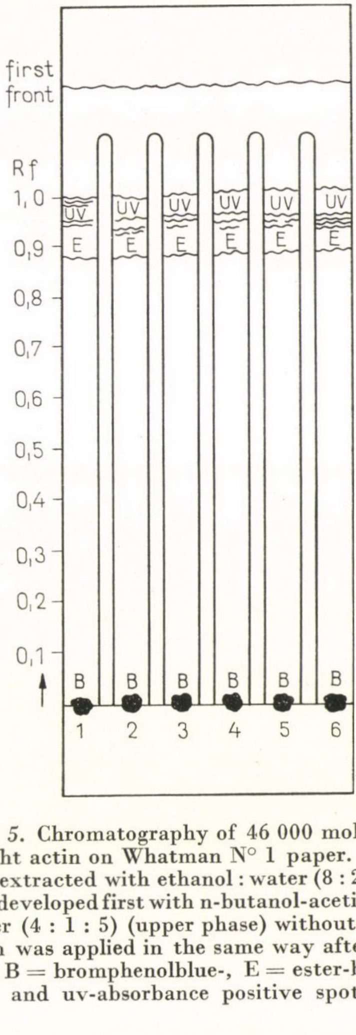

Fig. 5. Chromatography of 46 000 molecular weight actin on Whatman N° 1 paper. Paper was extracted with ethanol : water (8 : 2, v/v), and developed first with n-butanol-acetic acid-water (4 : 1 : 5) (upper phase) without actin; actin was applied in the same way after drying. B = bromphenolblue-, E = ester-bound- and uv-absorbance positive spots

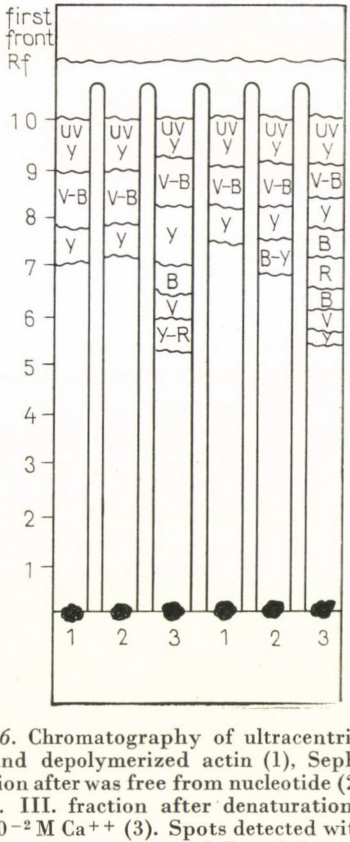

Fig. 6. Chromatography of ultracentrifuged, re- and depolymerized actin (1), Seph. III. fraction after was free from nucleotide (2), and Seph. III. fraction after denaturation with 5×10^{-2} M Ca^{++} (3). Spots detected with ninhydrin spray reagent followed heating at 80 °C. Spots are: Y = yellow, V-B = violet-blue, R = red, Y-R = yellow-rose, according to different lipid compounds (Hu *et al.* 1965). Other legend as in Fig. 5

Protein can be detected at the place of application even in the case of ultra-violet radiation and bromo-phenol-blue staining, while along the front line and near to it ester-type, acidic yellow lipid components are found. Chromatograms run parallel are made visible by ninhydrin staining, and then in the last quarter of the limit phase (front) violet, red, blue, yellow spots of unidentified components of lipid nature appear (Fig. 6).

Lipid extraction with butanol. According to our observations actins overtreated with the Dowex 50 ion exchange resin contain the lowest amount of molecule-bound components of non-protein nature. These actins were subjected to butanol extraction by the method of ZWAAL—VAN DEENEN (1968). The actin solutions were extracted four times with 1/4 vol. n-butanol, then after standing for one or two hours centrifuged. In the case of the butanol treatment three phases are obtained. After concentration the upper butanol phase contains 9.4 per cent of the initial material, while the interfacial layer 13 per cent. Here the protein fraction was determined to be 78 per cent, while on the basis of optical density it was 82—87 per cent. The above can be seen in Table 5.

Table 4

Nucleotide-, phosphorus- and lipid content in actin preparations obtained by various methods

Actin fractions	Number of isolat- ion	Total P-content**		Lipid content %	Phospho*-lipid content M/46 000 g	Protein P-content M/46 000 g	Nucleotide content M/M
		M/60 000 g	M/46 000 g				
Polymerized, purified once	4	9—13		15—20		1.0—1.5	0.8—0.9
Martonosi	10	6—7	4.9	13—15	0.5—1.5	0.5—1.2	0.8—1.0
Dowex treated	6	3.7—4.5	2.9—4.5	7.10	0.5—1.1	0.3—0.7	0.87—0.95
Myofibrillar	3		6.9	12—20	0.35—0.5	5.5—6.7	
Dystr. myofibr.*** actin	2		6.9—7.6	11—20	0.5—0.9	6.0—7.0	
Gel filtrated actin	5		3.5—4.5	10	0.4—1.0	0.4—0.9	0.35—1.0

* Phospholipid content in total lipid obtained by three times repeated extraction.

** Conversion of phosphorus content in actin obtained from purified myofibrillar suspension into 46 000 g molecular weight was made arbitrarily for the sake of comparison Conversion into 60 000 g molecular weight reflects the molecular weight accepted several years ago.

*** Myofibril prepared from dystrophic muscles of rabbits suffering from E-avitaminosis.

Table 5

Fractions of actin purified by Dowex II and -50 treatments after n-butanol extraction

Number of experiment	Butanol fraction %	Interfacial lipoproteid %	Lower aqueous weight %	Protein fr. E_{280} %
3	9.4/7.1—13	13	78 (68—85)	82—87
	organic acids	lipid + protein	protein + lipid	

After vacuum concentration the butanolic fraction is yellow and contains ester bonds and phosphorus besides the fatty acids.

The protein part of the interfacial lipoproteid is a single component (30 per cent), but the lipid fraction is a mixture (70 per cent, Fig. 7).

On a Dowex 50×2 column the fraction eluates in five fractions.

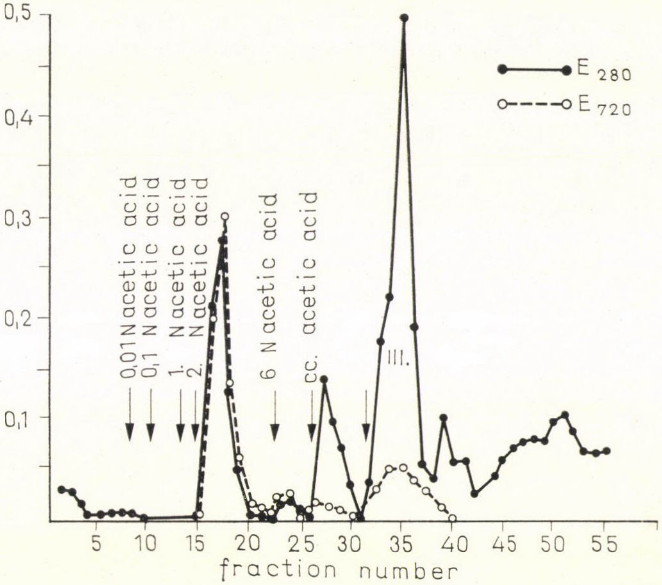

Fig. 7. Chromatography of interfacial lipoprotein by Dowex 50×2 column. The column was equilibrated with 0.1 N acetic acid-Na-ac buffer (pH 6.4), 40 mg lipoprotein was applied on the column. Elution as shown on the chromatogram. Fraction from peak III exhibited biuret positive reaction and typical protein uv spectra, while others flat spectra

The highest peak, fraction IV. is protein/biuret positive, the rest is biuret negative/, and are in the following order on TLC-plate:

fr. I. (15—20)	free fatty acid
fr. II. (22—26)	phosphatidic-acid, diphosphatidyl-glycerol
fr. III. (26—30)	phosphatidyl serine, ceramide, amino-ethyl-phosphatide (in traces), phosphatidyl-cholin
fr. V—VI. (38—?)	mixture; spots corresponding to the former fractions, and phosphatidyl-inositil, sphyngomyelin.

Lipid content in gel filtrated actin preparations. In spite of the removal of lipids the actins contain nonprotein substances. It was therefore considered reasonable to perform the gravimetric determination of gel filtrated actin preparations as well, although the other matters found beside the protein were not necessarily of lipid nature. The dialyzed, gel filtrated actin was dried in a vacuum thermostat at 105 °C, and gravimetrically measured, then the protein extracted three times with a mixture of chloroform-methanol-water (65 : 33 : 2, v/v/v). One homogenization and one extraction was carried out each day. It was concentrated from the organic solvent at 40 °C under an N atmosphere, and the residue as well as the lipid extracted protein measured. Lipid content amounted to 10—12 percent of the initial material dried at 105 °C (with a molecular weight of 46 000 g taken into account some 5000 g lipid extract was obtained).

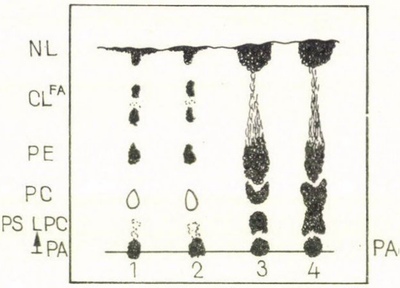

Fig. 8. Thin-layer chromatography of myofibrillar lipid by using silica gel. 1,2 are developed immediately after isolation, and 3,4 after autooxidation of lipids with Chl—MeOH—25%NH_3 (17 : 7 : 1, v/v/v) by Cuzner—Davison. PA = phosphatidic acid, PS = phosphatidyl serine, LPC = lysophosphatidyl cholin, PE = phosphatidyl ethanolamine, CL = cardiolipin, FA = fatty acids, NL = neutral lipid

In the course of IR spectroscope examinations these components show an absorption spectrum against chloroform. The substances show IR maxima characteristic of esters at the following Vmax/cm^{-1} values:

2962, 2862, 1465	(characteristic of —CH_3)
2926, 2853	(characteristic of —CH_2)
1720, 1740	(characteristic of ester bond)
1270—1290, 1250, 1180	(characteristic of unsaturated esters)
lower maximum 1570	(characteristic of amide bond)
lower maximum 1050—1080	(characteristic ether bond) and one
lower maximum 880	(characteristic of peroxide bonds)

occur as soon as 1—2 hours after isolation. Other absorption maxima are missing. The 963 cm^{-1} absorption band characteristic of trans double bands is missing. Thus, the substances obtained are supposedly fatty acid- and glyceride-, and perhaps phospholipid derivatives.

Finally, let us compare the thin-layer chromatograms of lipids obtained from myofibrillar suspensions and actin preparations.

Myofibrillar lipids were found to contain six components when freshly isolated and run, and fewer components when autooxidized (Fig. 8).

Gel filtrated actin showed three components when fresh, and five components when obtained from a dialysing liquid and autooxidized (Fig. 9).

Lipids obtained from dialysing solutions show perfectly different localization after autooxidation (Fig. 10).

The chromatograms presented do not show the complete picture, inasmuch as lipids obtained by extraction from myosin as well as those obtained from dialysing solutions are discussed in the next publication. Fraction Seph. I. contains mainly lecithin — left out as a contamination accompanying the actin —, although it is probably one of the lipid components of myofibril.

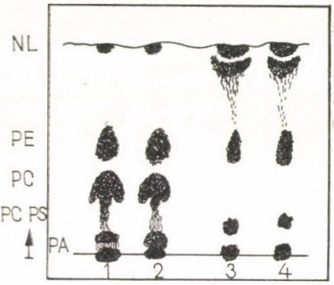

Fig. 9. Thin-layer chromatography of lipids from gel filtrated actin with Chl—MeOH—25% NH_3 by Cuzner—Davison (1966). 1.2 Lipids obtained from dialyzed actin under nitrogen atmosphere, 3.4 autooxidized lipids isolated from dialyzing solutions. Spot marks as in Fig. 8

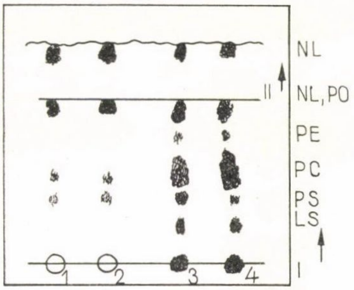

Fig. 10. Thin-layer chromatography of actin lipids, developed by Skipski *et al.* (1963). I Chl—MeOH-acetic acid—water (50 : 25 : 7 : 3, v/v/v/v), II n-hexan-diethyl ether-acetic acid (90 : 50 : 5, v/v/v). 1.2 Lipids from dialyzed actin after autooxidation, 3.4 Lipids from dialyzing solution. Spot marks as in Fig. 8

Discussion

In our investigations we found that the highest amount of phosphorus was contained by the purified myofibrillar suspension (an average of 21.5 g-atom/g). Only a minor part of the phosphorus content belonged to the nucleotides (1.5—1.8 micromol nucleotide/g). After the nucleotides had been removed a large amount of phosphorus was still left behind in the myofibril (17.1 g atom/g).

Further quantities of phosphorus contained in the myofibrillar suspension could only be removed partly by extraction performed on three successive occasions. Folch's lipid solvent mixture is able to remove 11—20 percent of the myofibril. This value may be most in vivo, since washing and dialysis against a buffer remove biuret negative components from the myofibril, moreover, some biuret negative substance can be obtained in this way even from the lipid extracted myofibrillar residue.

Only some 10—12 per cent of fractions obtained with a lipid solvent is phospholipid, which is slightly more than 1 percent of the dry matter content of myofibril. By similar methods BIRÓ—MÜHLRÁD (1960) could only remove 5 per cent of the inorganic phosphorus content of myofibril, while in the case of 6 per cent perchloric acid some 18 percent in four days (MÜHLRÁD *et al.* 1964).

Among the data concerning the actin only the gel filtrated actin is dealt with here. Investigations on actins obtained by the methods described in the experiments (cyclic precipitation, etc.) indicate heterogeneity, just like gel filtration itself. However, the fact that from gel filtrated actin some 10 per cent (4—5 mg/46 mg actin) pale yellow oil can be obtained, cannot be left out of consideration. Namely, with molecular rations taken into account, they may represent significant quantities in the native actin.

On TLC plate myofibrillar lipids show at least six spots, while the lipids of actin only four. In myofibrillar lipids developed with resorcin another spot can be demonstrated above the spot of lecithin, unlike the actin. The lecithin fraction of actin almost completely disappears after autooxidation, and instead of it 1—3 new spots appear near the solvent front, as well as a spot corresponding to hydroperoxids at the Rf-values of 0.11—0.15. The presence of unsaturated lipids may determine the tertiary and quarternary structures of structural proteins, and their rapid autooxidation may have a role in the heterogenization of actin and modify the denaturation of protein by establishing specific bonds between the polypeptid chains through the activation of double bonds. It may be responsible for the hypochrome effect of actin as well as for the fact that during the paper chromatographic separation of non-gel filtrated and gel filtrated actions the chromatogram becomes ninhydrin-positive owing to oxidation products of various degree and keto-groups formed as a result of lipid autooxidation. HU *et al.* (1965) found that keto-groups are stained different shades of colour when examined by paper- and layer chromatography, respectively. The small quantities of lipids present in the purified preparations can easily be believed contaminations, as they perhaps were considered to be by those noticing them during preparation and paying no special attention to their presence.

Acknowledgement

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ANATOMY OF VEGETATIVE FOOD STORAGE ORGANS

II. STEMS

By

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The anatomy of the vegetative food storage organs of *Amorphophallus campanulatus* and *Colocasia esculenta* has been investigated. Both of them have thick periderm. The massive ground parenchyma have few, scattered vascular bundles, a large quantity of starch grains and numerous isolated laticifers. Their structural modifications in relation to the function they perform are discussed.

Introduction

We attempted to elaborate in the first article of this series the structural organization in the food storage organs which appears to be directly related to the function they perform. Furthermore, although it may vary from plant to plant, most of the manifestations are on the same line. To determine these modifications the anatomy of certain food storing roots was studied and it was revealed that although they perform a similar function and possess an identical ground plan, they have a variable organization which appears to be controlled by the genetic make up of the species. The present investigation is an extension of the work on roots referred to earlier (PALIWAL—KAVATHEKAR 1971).

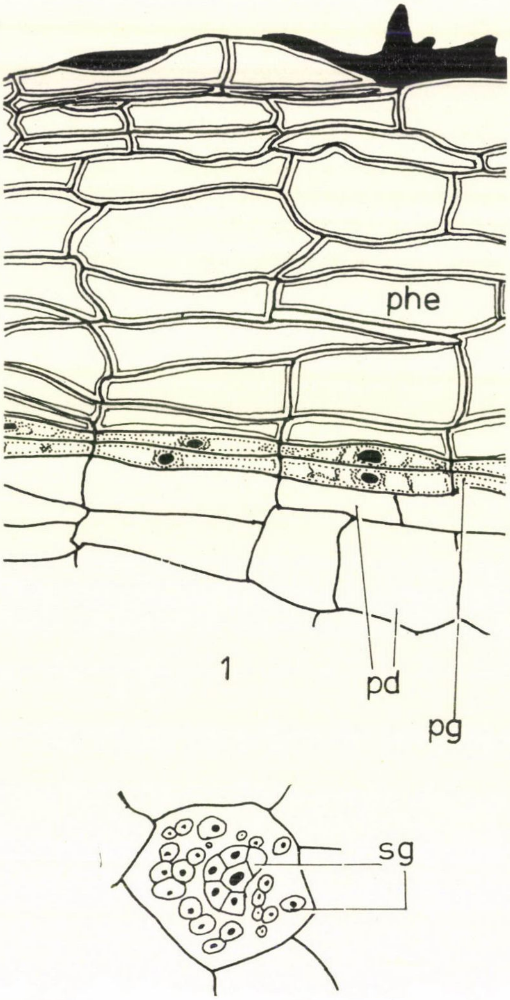
Material and Method

Amorphophallus campanulatus (Roxb.) Bl. ex. Decne. (vern. name Zaminkaṇd) and *Colocasia esculenta* (Linn.) Schott. (vern. name Arvi) both belonging to the family Araceae were chosen for the study. The edible part was obtained from the local market. It was chopped up into small bits (about 1—2 cms each), fixed in FAA and stored in 70% ethanol. Sections were cut with the help of a sliding Jung microtome at a thickness of 25 to 40 μ. These were stained with safranin-fast green combination and with iodine alone.

Results

Amorphophallus campanulatus

The fleshy, edible part is a corm — an underground modification of the stem. It is generally hard, bulky and dark grey coloured. Being a monocot the internal structure of the organ is very simple. The periderm covers the entire body and is continuous throughout. The cork is 5 or more layered (Fig. 1). It comprises rectangular to hexangular, thick-walled cells placed one above the other in a rather loose manner. These have poor cytoplasmic contents, the latter being located only in the peripheral region. The cork cambium is clearly distinguishable and consists of one to three layers of thin-walled, nucleated, rectangular cells. The cortex or ground parenchyma (Fig. 5) usually comprises circular cells but these may have variable shapes also. Laticiferous tubes occur scattered. As seen after staining these are dark coloured, possess



Figs 1—2. Periderm and starch grains of *Amorphophallus campanulatus.* (*phe,* phellem; *pg,* phellogen; *pd,* phelloderm; *sg,* starch grain). — 1. Periderm t.s. ×400. — 2. A parenchymatous cell with starch grains. ×400

dense contents and generally occur singly. At various places the ground parenchyma is aggregated into regions containing starch grain and regions without, the former staining darker than the latter. The cells of the light-stained areas may also possess a few starch grains.

The starch grains are circular and may occur isolated or in groups (two to many; Fig. 2). The cells of the dark stained regions are packed with starch grains having a central hilum. These cells also contain many fat globules besides the starch grains. Ground parenchyma is rich in druses and crystals of various shapes.

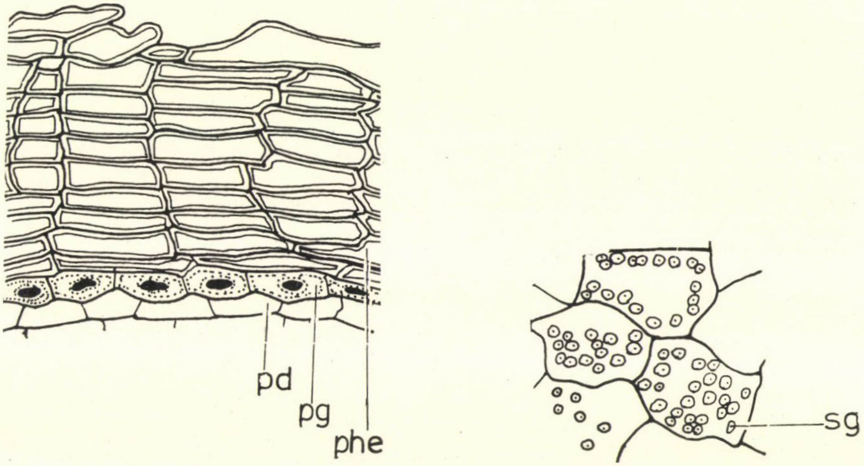
The vascular bundles are scattered, few in number and lack cambium.

Colocasia esculenta

Food is stored in the modified stem which is underground and is described as a rhizome. It has various shapes and sizes and may be round, oval to elongated, or even rod shaped. It is generally light green to dark brown in colour and is covered with a thich blanket of scaly leaves.

From its very early stages of development, the rhizome is covered completely by a thick periderm and in the mature structure as many as 24—30 cork layers may occur. The cork cells are rectangular, delicate with little thickening and arranged in parallel lines one above the other (Fig. 3). The cells have scanty cytoplasm.

The ground parenchyma, located just below the cork cells also shows the presence of cork cambium (1—2-layered). The cork cambium is followed



Figs 3—4. Periderm and starch grains of *Colocasia esculenta. (phe,* phellem; *pg,* phellogen; *pd,* phelloderm; *sg,* starch grains). — 3. Periderm in t.s. ×400. — 4. Few parenchymatous cells with starch grains. ×400

Table 1

Comparison of anatomical features of the vegetative food storage organs of A. campanulatus and C. esculenta

Character	*A. campanulatus*	*C. esculenta*
1. Cork	*a)* 5-or more layered *b)* recta-, penta- or hexangular in t.s. *c)* not arranged in tiers	*a)* 25—30 layered *b)* only rectangular in t.s. *c)* arranged in tiers
2. Mucilage ducts	absent	present always form a ring below the cork cambium
3. Latex cells	scattered, unbranched and short	scattered, branched, very long and generally associated with vessels
4. Starch grains	*a)* in two conspicuous regions *i)* cells rich in starch grains *ii)* cells poor or completely lacking starch grains *b)* hilum distinct	*a)* present uniformly *b)* hilum insignificant

These differences are probably due to the genetic differences of the two plants.

by a ring of isolated mucilage ducts of various sizes which are produced lysogenously. The mucilage secreted by these ducts makes the rhizome very slippery especially if touched after peeling off the periderm. The ground parenchyma (Fig. 6) cells are small and variously shaped. The starch grains (Fig. 4) are uniformly distributed all over the rhizome. The former are more or less circular with an insignificant hilum.

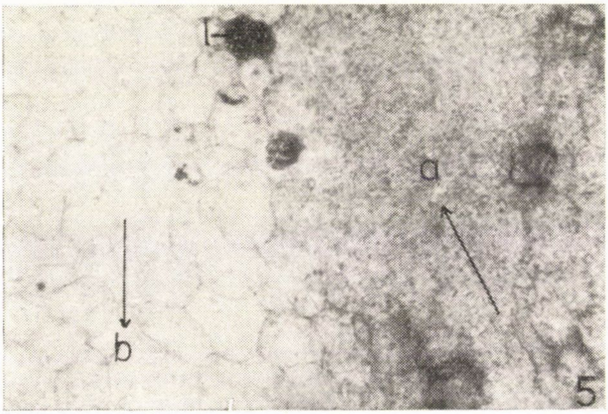


Fig. 5. Ground parenchyma of *A. campanulatus (a,* region with cells rich in starch grain; *b,* region with cells with fewer starch grains; *l,* latex cell) × 353

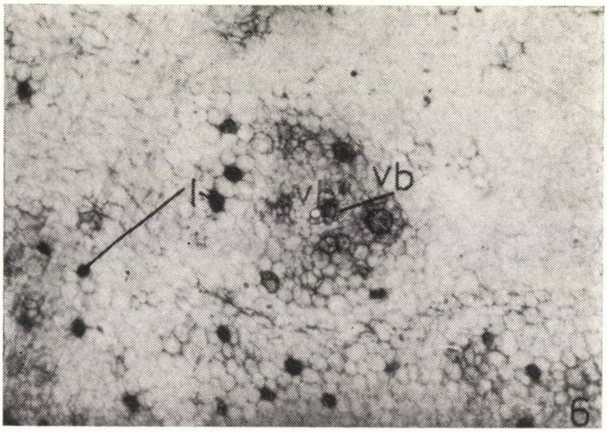


Fig. 6. Ground parenchyma of *C. esculenta* (*l*, latex cell; *vb*, vascular bundle). ×353

The ground parenchyma is characterized by the presence of a large number of isolated, individual laticifers. One laticifer extends to as many as 20 parenchyma cells in longitudinal section and may be branched.

Discussion

The characters which are reported to be common to the other food storage organs such as in *Solanum tuberosum* (Artschwager 1924); *Beta vulgaris* (Artschwager 1926); *Daucus carota* (Esau 1940); *Ipomoea batatas* (Govil 1969); *Brassica rapa*, *Dioscorea bulbifera*, *Ipomoea batatas*, *Manihot esculenta* and *Raphanus sativus* (Paliwal—Kavathekar 1971) are worthy of mention: 1. An abundance of periderm which is predominated by thick-walled cork cells arranged in tiers. 2. The food storing capability is best carried out by the large, thin-walled parenchymatous cells lacking intercellular spaces. 3. The vascular bundles are relatively poorly developed and are fewer in number. 4. Latex cells, mucilage cells can be found along with ergastic substances such as druses and raphides.

The characters observed in *Amorphophallus campanulatus* and *Colocasia esculenta* although both are stems follow the same pattern of organization as have been reported in the above plants all of which are the roots modified to take this function of storage except *S. tuberosum*.

Although both the genera belong to the same family, perform a similar function and possess an identical ground plan they have some characters quite different from each other.

Acknowledgements

We are grateful to Professor B. M. Johri for facilities and encouragement.

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CHARACTERISTICS OF THE MEMBRANE AT THE POLLEN PORES OF SOLANUM DULCAMARA L. I

DISINTEGRATION OF THE MEMBRANE

By

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According to researches the cytoplasmic trabeculae interweaving the intine at the pores of the pollen grains as well as their parts protruding from the intine are covered by a plasma membrane. This supposition has been confirmed by our observations made on the pollen of *Solanum dulcamara* L. namely: if calcium ensuring the stability of this membrane is extracted from the membrane structure by compounds binding the calcium, then the stability of the membrane will cease. In this case the membrane disintegrates and the cytoplasm of the vegetative cell flows away through the pores. Besides proving the existence of a membrane structure at the pores, our observations have practical importance too. At the time of flowering, and when the pollen grains scatter from the anthers, no plant protectives should be used which by getting into the plasma membrane below the intine at the pores bind the calcium which ensures its stability. Namely, the pollen grain whose plasma membrane at the pores disintegrates, and the cytoplasm of whose vegetative cell flows away through the pores will neither be able to develop a tube nor fertilize.

Introduction

Below the pores of the pollen grains only the intine is continuous. Otherwise, on the parts adjacent to the pore five layers (intine, endonexine, ectonexine, endosexine, ectosexine) can be distinguished of which the two nexines and sexines form a fine reticulum. At the pores the intine is densely interwoven by thin cytoplasmic trabeculae perpendicular to the plane of the intine, which reach to the outer surface of the intine or even emerge from there. On the basis of researches on fine structure it was supposed that these cytoplasmic trabeculae and their parts rising from the intine were also covered by plasma membrane. This membrane is similar to or identical with the plasmalemma surrounding elsewhere the cytoplasm of the vegetative cell, and is its direct continuation, respectively (Erdtman 1952, Mühlethaler 1955, Ledbetter—Porter 1970). Inasmuch as these membranes surrounding the cytoplasmic trabeculae were plasma membranes and the continuations of the plasmalemma, the characteristics of the outer plasma membranes of plant cells would be easy to study without any technical intervention at the pores of the pollen grains.

One of the important components of plasma membranes is the calcium which takes part in building up the structure of the membrane, determines its quality (WEIGL 1967), has a role in producing the membrane potential (HIGGINBOTHAM *et al.* 1964, KISHIMOTO 1966a, b, c), but its main task is to stabilize the membrane structure. If the quantity of calcium contained in the plasma membrane is reduced by means of compounds binding the calcium, then its ion accumulation capacity decreases (HERMAN 1964). In case of a total absence of calcium the cytoplasm flows away, since the stability of the plasma membrane has ceased, and the cell dies (DE ROBERTIS—NOWINSKI—SAEZ 1970).

We started from the hypothesis that — according to the researches on fine structure — the cytoplasmic trabeculae interweaving the intine, and their protrusions too are covered by plasma membrane. This plasma membrane is the direct continuation of the plasmalemma of the vegetative cell. The stability of plasma membranes is ensured by calcium. Thus, if the calcium is extracted from the membrane at the pore by means of compounds binding the calcium, the membrane necessarily disintegrates, and the cytoplasm of the vegetative cell flows away through the pores of the pollen. This phenomenon can be well seen through a standard microscope.

Material and Method

The material of our investigations was mature pollen from the flower of *S. dulcamara* L. which we treated with the following solutions containing calcium binding compounds: $(COOH)_2$, Na_2HPO_4, Na_2CO_3, $K_4[Fe(CN)_6]$, 0-oxichinolin.

From $(COOH)_2 \cdot 2\,H_2O$ a 1 per cent solution was prepared with distilled water; from the reagent containing Na_2HPO_4 similarly a 1 per cent solution; a 1 per cent solution was first made from the Na_2CO_3 reagent too, but no kind of effect was observed even after some time of standing, therefore the concentration of the reagent solution was raised to 5 per cent; from the reagent containing $K_4[Fe(CN)_6]$ slightly ammoniacal neutral solutions of various concentration from 1 per cent on; in the same way, from 0-oxichinolin slightly ammoniacal alkaline solutions of various concentration from 1 per cent on were prepared.

One drop of each of the above solutions was mixed separately on a slide with two drops of 50 per cent saccharose solution. In this way the concentration of the solution became identical with the osmotic concentration in the vegetative cell of the pollen grain. The solution was spread over the slide and sprinkled evenly with pollen from the mature anther of *S. dulcamara.*

Preparations thus prepared were placed in Petri-dishes on cotton-wool saturated with hot water, in order to provide an atmosphere of saturated vapour content for the pollens after the Petri-dishes had been covered. Then the Petri-dishes were placed in a thermostat at 30 °C, and the preparations examined every half hour through a Leitz Ortholux microscope.

With each reagent more than one parallel tests were performed simultaneously, and the tests repeated several times. So our results were obtained by examining several thousand pollen grains.

Results

In the course of evaluating our experiments we examined the changes through a standard microscope that solutions of various calcium-binding compounds brought about at the pores of the pollen grains. Namely, if the reagent employed gets into the plasma membrane — inasmuch as it is a membrane — and binds the calcium that ensures its stability, then the stability of the membrane ceases. The membrane disintegrates and the cytoplasm of the vegetative cell flowing away through the pore will be visible through a standard microscope.

In the case of treatments with $(COOH)_2$, and Na_2HPO_4 in the preparation examined through a microscope the stability of the membrane covering the cytoplasmic trabeculae interweaving the intine at the pore was found to have ceased already after half an hour. The membrane disintegrated and the cytoplasm flowed away through the pore. After an hour this phenomenon occurred in each pollen grain. In the case of multipore pollen grains, such as those of *S. dulcamara*, the plasma membrane disintegrates and the cytoplasm flows out first at only one of the pores. Later, however, this phenomenon takes place at the other pores too.

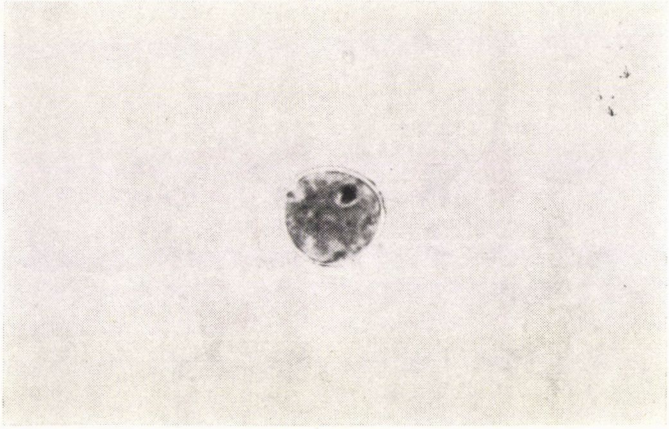

Fig. 1. No change as a response to carmine acetic acid staining can be found

In the case of the reagent Na_2CO_3 disintegration of the plasma membrane and flowing out of the cytoplasm occurs only when higher concentrations are used, and then more slowly. Flowing out of the cytoplasm is preceded by its becoming highly granulated (Figs 1 to 6).

We consider the phenomenon of the cytoplasm flowing away through the pores under the influence of treatments with $(COOH)_2$, Na_2HPO_4 and Na_2CO_3 to be an evidence that the cytoplasmic trabeculae interweaving the intine and protruding from it are covered by plasma membrane. This plasma membrane is the direct continuation of the plasmalemma in the vegetative cell. In this case the cytoplasm of the vegetative cell and the plasmalemma covering it reach into the close-mashed screen-like holes of the intine at the pore and communicate directly with the outer environment.

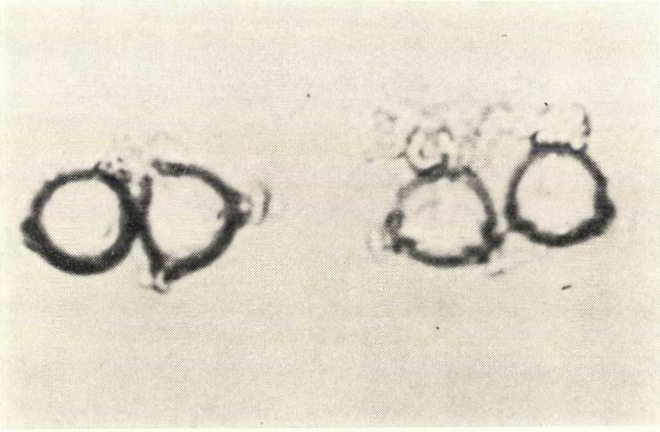

Fig. 2. Under the influence of $(COOH)_2$ the cytoplasm of the vegetative cell flows away through the pore

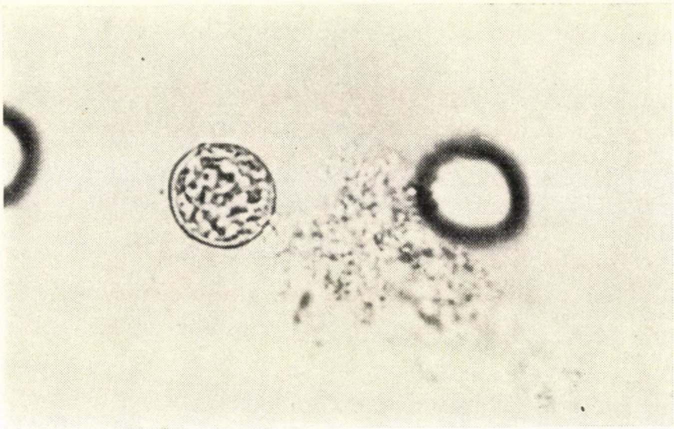

Fig. 3. Under the influence of Na_2HPO_4 the cytoplasm of the vegetative cell flows away through the pore

Treatments with $K_4[Fe(CN)_6]$ and O-oxichinolin. After preforming treatments with the two reagents separately we did not find changes in the preparations examined through a microscope, either when increasing the concentration of the solutions of when prolonging the time of standing of the preparations. The stability of the plasma membrane remained unchanged, it did not disintegrate, consequently the flowing away of the cytoplasm could not be observed during the microscopic studies when several thousand preparations were examined.

The reason why treatments with $K_4[Fe(CN)_6]$ and O-oxichinolin did not result in the same phenomenon as the other calcium-binding compounds was

Fig. 4. Under the influence of Na_2CO_3 cytoplasm becomes first granulated

Fig. 5. Under the influence of Na_2CO_3 later the cytoplasm of the vegetative cell flows out through the pore, or flows away

— in our opinion — either the fact that the molecules of the reagent could not get to the calcium in the membrane, or the stability of the complex ions, that is the dissociation of the component into ions (second dissociation).

Pollen tube development. The pollen grains did not develop tubes either when the plasma membrane disintegrated at the pores or when it remained intact. In culture media with similar sugar concentration, and under the conditions of identical relative humidity, similar temperature and the same period of time, tube development in all cases occurred if the culture medium did not contain calcium-binding substances.

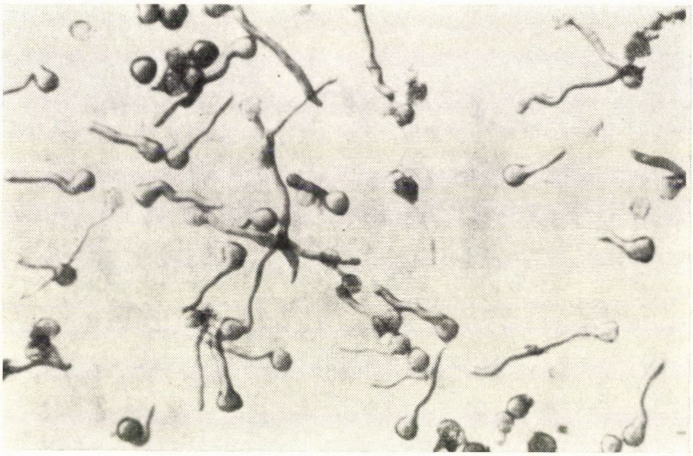

Fig. 6. On culture media not containing calcium-binding substances the pollen grains develop tubes

Disintegration of the membrane structure at the pore proves that the plasmalemma of the vegetative cell is in direct connection with the outer environment, while the failure of pollen grains to develop tubes is the evidence of environmental factors determining the tube development.

Discussion

On the basis of researches made on the fine structure it was supposed that the cytoplasmic trabeculae interweaving the intine at the pores of pollen grains as well as their parts protruding from the intine were covered with a plasma membrane. The membrane separating the cytoplasmic trabeculae and their protrusions from the pore disintegrate under the influence of substances binding the calcium ions, and the cytoplasm flows away through the pore. This proves the unity of the membrane structure on one hand, the direct contact of the plasmalemma in the vegetative cell with the outer environment on the other, and finally the influence exercised by the outer environment on pollen tube development. Besides all this the above statement has practical importance too. At the time of flowering and when the pollen grains scatter from the anthers, plant protectives which by getting into the plasma membrane through the pores bind the calcium which ensures its stability — should not be used. Namely, the pollen grain whose plasmalemma disintegrates at the pore and the cytoplasm of whose vegetative cell flows away through the pore, is no longer able either to develop a tube of fertilize.

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EFFECT OF CCC TREATMENT ON VARIOUS STONE-FRUIT SEEDLINGS

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The author applied CCC treatments to wild cherry- and mahaleb seedlings at concentrations of 0, 500, 1000, 2000 and 4000 ppm and to myrobalan- and wild apricot seedlings at 0, 2000 and 4000 ppm. Permanent growth inhibition was observed only in varieties of high growth vigour (wild cherry and myrobalan). The author further pointed out a positive correlation between the inhibition of internode elongation and the incresase of crude protein content.

Introduction

The growth inhibition of shoots caused by (2-chloroethyl) trimethylammonium chloride (CCC) opens up broad vistas in fruit production. CCC treatments proved especially efficient in the case of pear, cherry, currant and strawberry (Góra 1968, Sági—Bubán—Zatykó 1970) while had hardly any effect on apricot (Vavra—Musoliva 1968).

Generally, growth retardants increase the total N-content of plants (Barbier—Mayr 1966), but in bean leaves they may reduce it (Humphries 1968). Under the influence of CCC the calcium content in the leaves decreases, at the same time, in the author's experiments it significantly increased in the woody parts of wild pear seedlings. A similar quantitative increase was observed in the total carbohydrate content (%) (Surányi 1970) as was demonstrated earlier with germinating wheat grains too by el-Fouly—Jung (1966).

In the author's present study the effect of CCC on seedlings of different growth vigour on one hand, and a possible correlation between induced growth inhibition and changes of calcium-, total carbohydrate- and crude protein contents in woody parts of plants, on the other, were investigated. The purpose of using seedlings as test plants in the experiments was to avoid the modifying effect of pruning on nutrient transport (Brunner 1968) and shoot growth (Flierman—Houter 1970).

Material and Method

Populations of wild cherry C. 2794, mahaleb C. 2742, myrobalan C. 359 and wild apricot C. 2700 were transplanted on 10 May 1969 from a seedling nursery into a greenhouse. Treatments began on 13 June: wild cherry and mahaleb were treated with CCC of 0, 500, 1000, 2000 and 4000 ppm concentrations, while the myrobalan- and apricot seedlings on 20 June with CCC-solutions of 0, 2000 and 4000 ppm. Treatments were performed on nine occasions with two days intervals in the form of painting the shoot-apices with the CCC-solution. No water was given to the plants on treatment days, only in the intervals. Plant heights were measured and leaves counted every two weeks from the beginning of the treatment.

During the experiment stem elongation was expressed for each plant (10 seedlings per treatment) as a percentage of the initial plant height; when the measurements were completed the average length of internode was calculated from the length of the stem and the number of leaves. The results obtained were evaluated by variance analysis. After the removal of the leaves the woody parts were chopped up, then, when dried, the material was ground, the Ca-, total carbohydrate- and crude protein content determined and statistically evaluated (2 replications).

The Ca-content was determined according to the standard method MNOSZ 448-55. 1 g of wood dust was reduced to ashes by heating at 800 °C over two and a half hours, after being dissolved in hydrochloric acid the remaining part was treated in the same way as prescribed for the normal water Ca-content determination. Finally, the solution neutralized with NaOH of 33 per cent was titrated with Complex III while using Murexid as an indicator.

For the total carbohydrate determination the GOSZT 559 standard was used in a modified form. 2.5 g wood dust was washed with warm water into a 200 ml volumetric flask and — after 15 ml of 20 per cent hydrochloric acid had been pipetted to it — boiled for 3 hours. When the solution had cooled it was neutralized approximately by NaOH of 10 per cent concentration, then diluted with distilled water to 20 ml. After filtration, 50 ml of the filtrate was neutralized perfectly in the presence of methylene orange indicator and 4 ml of both zinc acetate and potassium ferrocyanide were added to it. After dilution the filtration was repeated. The end-filtrate was titrated with Fehling's solution in the presence of a methylene blue indicator.

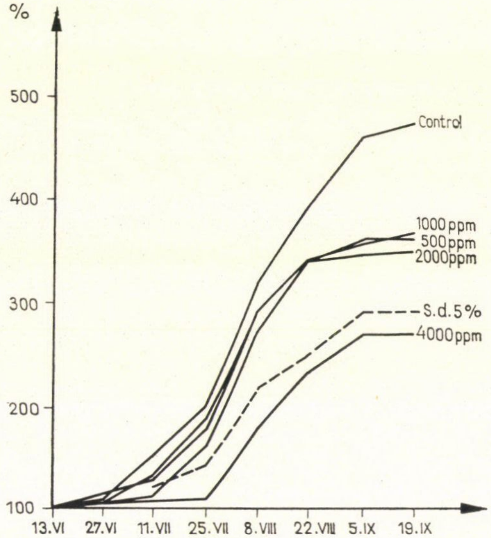
The crude protein was determined on the basis of the MNOSZ 6830-53 standard. Wood dust dried at 105 °C was destroyed in a 300 ml Kjeldahl flask over 2—3 hours until it was completely bleached. A mixture of potassium sulphate and copper sulphate at a ratio of 4 : 1 as well as some selenium were used as catalyzers. When the solution had cooled it was completed to 100 ml then distilled. A 10 ml quantity of the developed material was pitepted into a Wagner—Parnas destilling flask, and the ammonia distilled through hot steam, with an excess of 33 per cent NaOH into an Erlenmeyer flask. Distillation lasted for five minutes. 10 ml 0.1 per cent hydrochloric acid stained with methylene orange was used as recipient fluid. The solution was then retitrated with 0.1 n NaOH until the methylene orange assumed a transitional colour.

Results

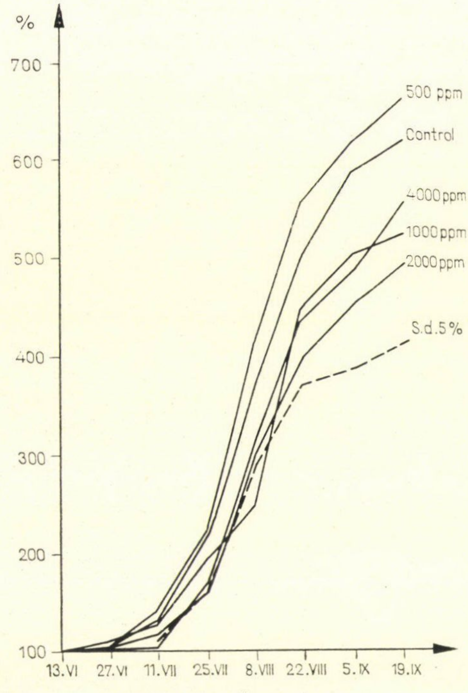
The CCC-treatments caused different degrees of growth inhibition in wild cherry- and mahaleb seedlings. Growth inhibition caused by a concentration of 4000 ppm in wild cherry was of such an extent that the seedlings could not overcome it in the rest of the vegetative period. The other concentrations had hardly any effect on the plants. In the mahaleb seedling the influence of CCC was extremely low, only the 1000 ppm concentration caused a temporary inhibition as compared to the control (Fig. 1).

The effect of CCC on myrobalan — and wild apricot seedlings was similarly different. The myrobalan seedlings could not outgrow the effects of 2000 and 4000 ppm treatments by the end of the vegetative period. Essentially the wild apricot seedlings did not give a striking response to the chemical (Fig. 2).

Table 1 shows that in seedlings which displayed the effects of CCC-treatments even at the end of the vegetative period the average length of the inter-



a



b

Fig. 1. **Effect of various concentrations of CCC on stem elongation in wild cherry C.2794 (A) and mahaleb C. 2742 (B) (on 13th June 100 per cent). (Mean values below the broken line representing S.D.$_{5\%}$ differ significantly from the control)**

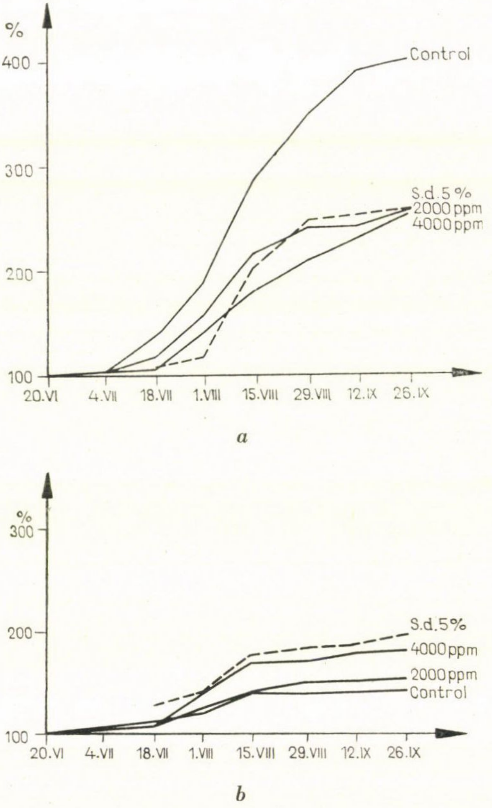


Fig. 2. Effect of various concentrations of CCC on stem elongation in myrobalan C. 359 (A) and wild apricot C. 2700 (B) (on 20th June 100 per cent). (Mean values below the broken line representing S.D.$_{5\%}$ differ significantly from the control)

nodes was behind that of the control. In the next column of the Table the Ca contents of the woody parts do not show unambiguously whether the efficient CCC concentrations also involve changes in the Ca content, and even the results significantly deviating from the control fluctuate in both directions (Ca content was increase by 500 and 1000 ppm in wild cherry and by 2000 ppm in wild apricot, while decreased by 500 ppm in mahaleb). Almost the same stands for the total carbohydrate content: in mahaleb- and myrobalan seedlings the 4000 ppm concentration invariably increased the total amount of carbohydrates, while in the woody parts of wild cherry- and wild apricot seedlings it showed a tendency to decrease under the influence of the same concentration of CCC. On the other hand, in the case of both permanent and temporary growth inhibition caused by CCC at concentrations of 1000 ppm in mahaleb, 2000 and 4000 ppm in wild cherry and myrobalan, the crude protein content showed an increasing trend regarding its percentage proportion to total dry matter.

Table 1

Effects of various concentrations of CCC on wild cherry- and mahaleb, myrobalan- and wild apricot seedlings

Treatment	Length of internode mm	Ca content ‰	Total carbohydrate %	Crude protein %
Wild Cherry C. 2794				
Control	15.88	6.25	32.10	8.61
500 ppm	16.35	7.50*	31.80	9.05
1000 ppm	13.43	8.94***	33.30	9.26
2000 ppm	13.89	6.97	30.00	9.69*
4000 ppm	10,54**	5.72	30.45	10.16**
Mahaleb C. 2742				
Control	19.44	8.40	27.60	8.93
500 ppm	18.70	6.07**	30.00	7.95
1000 ppm	17.37	8.58	28.65	9.69*
2000 ppm	16.62	8.22	30.45	8.29
4000 ppm	17.21	8.40	30.90*	7.95
Myrobalan C. 359				
Control	16.86	6.08	27.60	7.95
2000 ppm	12.55*	7.15	28.65	8.62*
4000 ppm	12.82*	7.15	29.25	9.50
Wild apricot C. 2700				
Control	10.01	7.86	27.60	14.58
2000 ppm	9.78	9.47*	28.05	15.46
4000 ppm	10.25	9.29	27.45	15.02

* at 5% level
* at 1% level significantly differs from the control
*** at 0,1% level

Table 2 summarizes the correlation studies. A definite negative correlation was found between the average length of the internode and crude protein content, that is, accumulation increased in seedlings with internodes shorter than that of the control crude protein. Among the other components examined in the woody parts Ca-and crude protein contents showed a positive while total carbohydrate- and crude protein contents a negative correlation.

Table 3 shows the effect on CCC-treatments on stone-fruits (wild cherry, mahaleb, myrobalan and wild apricot) of different growth vigour by comparing the percentage stem elongation caused by the various concentrations (500,

Table 2

Results of regression-analysis between the Ca content (%), total carbohydrate (%) and crude protein (%) and the average length of internode (mm)

Length of internode and Ca content	$r = -0.1897$	
Length of internode and total carbohydrate	$r = +0.2234$	
Length of internode and crude protein	$r = -0.7751$***	$Y' = 18.79-0.61x$
Ca content and total carbohydrate	$r = -0.1892$	
Ca content and crude protein	$r = +0.6884$**	$Y' = 1.65+1.09x$
Total carbohydrate and crude protein	$r = -0.6308$**	$Y' = 29.27-0.65x$

** at 1% level
*** at 0,1% level
the correlation is significant

Table 3

Comparison of the efficiency of CCC in stone-fruits of different growth vigour
(Percentage elongation is expressed in comparison to the control)

Time	Growth vigour	
	Intensive	Poor
	Wild cherry	Mahaleb[1]
July 11	78.4	98.9
Aug. 8	73.9*	85.8
Sept. 5	72.7*	87.6
Oct. 3	71.4*	90.2
	Myrobalan	Wild apricot[2]
July 11	98.4	97.9
Aug. 15	77.8*	110.2
Sept. 12	65.9*	114.5
Oct. 10	63.7*	115.4

Note: 1. Mean value of 500, 1000, 2000 and 4000 ppm treatments
2. Mean value of 2000 and 4000 ppm treatments.
* At 5% level differs from the other column.

1000, 2000 and 4000 ppm in four replications; 2000 and 4000 ppm in two replications) to that of the control. The variance analysis gave significant differences: seedlings showing a higher growth vigour (wild cherry and myrobalan) were more responsive to the CCC concentrations examined than those of a lower growth vigour (mahaleb, and wild apricot).

The varying efficiency of CCC is worth being examined not only from the point of view of growth vigour but also for its causes. The percentage proportion of cortex + phloem is significantly lower in the roots of wild cherry and myrobalan than in those of mahaleb and wild apricot. This means that mahaleb and wild apricot have a lower growth vigour when compared to wild cherry and myrobalan (BRUNNER—NYUJTÓ—ANTONI 1968, SEBŐK—ANTONI 1969). In biochemical studies on apple rootstocks of different growth habit GUR—SAMISH (1968) found that in the phloem of poorly growing rootstocks the auxin-oxidase activity was much higher and, accordingly, the endogenous auxin level lower, suggesting a close correlation between auxin-oxidase activity and growth vigour. Thus, in the present case CCC was only able to stop the growth of the shoots in species with a high growth vigour. This seems to be confirmed by investigations made by BRUNNER—ANTONI (1970) who pointed out a higher auxin content in wild cherry as compared to sour cherry, and in myrobalan compared to wild apricot.

Investigations on the components of woody parts gave, in essentials, no unambigouus answer to the question whether CCC really promotes the fruit trees in becoming bearing. Namely, parallel with the fruit trees becoming bearing many authors found the carbohydrate content to increase (KOBEL 1954), at the same time an increased Ca accumulation could also be observed (PERFILEV 1962). CCC seems to disturb the calcium transportation, while calcium deficiency inhibits the carbohydrate transportation (JOHAM 1957). This — though manifest as a tendency — could not be proved by correlation calculations. Increase in the crude protein content could sooner be the cause of a quantitative decrease in the carbohydrate content (r = —0.6308), since the reduction of nitrates requires carbohydrates.

The above results have contributed to the determination of the plant species spectrum of CCC. Further investigations are required, however, to clarify the effect of CCC treatments on the organic matter transportation. Furthermore, neither is it unimportant to find out the extent of growth vigour at which plant growth can be inhibited, since — as seen in the case of wild apricot and mahaleb — the CCC may be ineffective.

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A METHOD FOR SEEKING THE MOST INFORMATIVE CHARACTERS

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When studying a number of characters changing more or less interdependently we can expect to eliminate some of them without losing considerable information. From many aspects such reduction in the number of characters is highly desirable. In spite of this fact no acceptable method of seeking for the unimportant variates has been found so far in the literature. The present paper discusses the basic principles of a possible method.

Introduction

When more than one elementary components of a subject are studied simultaneously they often turn out to change interdependently. For example: the various yield components of cereals under different treatments, the morphological features of the different varieties of a fruit species, yields of various plants on different soil types, the various characteristics of an ameliorated soil during the years, the value of constituents in some animal product, etc. — all change rather parallelly. The interdependence between the studied characters (variates) is in many cases so close that some of them practically contain all the information that could be obtained by examining all the characters.

In the vast literature on multivariate analysis no method of seeking for variates that can be discarded is found, as pointed out by **Kendall** (1957) — an excellent specialist of the subject — on page 70 of his monography. However, solving the problem is extremely important for two main reasons: *a)* to examine variates in a number higher than necessary superfluously increases the costs; *b)* in the case of many variates the interpretation of results is complicated, and — owing to the interdependences — may even be false.

The idea of a possible solution was raised in one of the author's previous papers (1971); it was this idea that led to the method described in the present paper. Only the theoretical construction and the algorhythm of the calculation are presented here without the mathematical exposition of the procedure. The significance test will not be touched upon either.

Materials and Methods

1. The variates examined will be denoted by $x_1, x_2, \ldots x_p$. These variates are to be replaced by the variates $y_1, y_2, \ldots y_r$, (of reduced number) chosen from the x's. Let us suppose that the stochastic relationship between the variate pairs is linear, if not this condition should be attained by transformations as far as possible, lest the application of the method should lead to false conclusions.

For the sake of comprehension the following concepts will be introduced:

subject system: $X = (x_1, x_2, \ldots x_p)$, the totality of variates to be examined (in short: subject)

informator system: $Y = (y_1, y_2, \ldots y_r)$, the ensemble of variates that replace the subject system (in short: informator)

inducator (ϑ): the variate on which the changes in subject variates are examined (e.g.: year, treatment, sample, etc.).

2. The task consists — in essentials — of finding an index number which expresses the degree of reliability to which the informator Y determinates the subject system X, if the change in the latter is "along" the inducator ϑ. If a suitable index number is found, we can start searching for system Y supplying just as much information about the subject X as the latter supplies about itself.

After theoretical considerations the following term seemed to be the most suitable for measuring the information of Y about X:

$$I(Y \xrightarrow{\vartheta} X) = \frac{1}{p} \sum_{k=1}^{p} R^2_{x_k(Y)} \tag{1}$$

where $R^2_{x_k(Y)}$ is the square of the multiple correlation coefficient between the variate x_k and the system Y (coefficient of determination), which is also calculated along the inducator ϑ. This index number is obviously between 0 and 1, specially: $I(X \rightarrow X) = 1 = 100\%$. If each of X and Y consists of a single variate, the right side of (1) will be reduced to their (simple) correlation.

3. There is no difficulty in computing the above formula with X, Y and ϑ given. Further on only the technics of computation will be simplified. In accordance with the problem let us suppose that the informator will be chosen from the subject: $Y = (x_{i1}, x_{i2}, \ldots x_{ir})$.

Computation will be easy if the correlation matrix $R = (r_{jk})$ of variates $x_1, x_2, \ldots x_p$ is constructed. This contains the matrix R_y of the informator variates too. For the computation of I only the matrices $\frac{1}{p} R^2 = (q_{jk})$ and $R_y{}^{-1} = (c_{jk})$ are needed. By a short formal transformation we obtain:

$$I(Y \rightarrow X) = \sum_{j,k=i_1}^{i_r} c_{jk}\, q_{jk}. \tag{2}$$

By means of this formula the information of all possible informators can be computed quite quickly, in the case of smaller problems (p = not more than 6) with a desk machine, while in the case of bigger problems with a computer. There is no need, in fact, to examine all the possible informators, conclusions can be made regarding the expectations of the individual informators by various considerations without calculations.

4. Any informator Y can be decomposed into "disjunct" components, $y_1^*, y_2^*, \ldots y_r^*$, say (which do not contain information about one another); y_1^* is a linear function of the informator-variates which contains maximum information on the subject system; y_2^* contains the maximum proportion of the remainder information, and so on. The information contents of the individual components are given by the latent roots of matrix $R_y{}^{-1} Q_y$, while the components themselves can be produced by the latent vectors of this matrix, where Q_y is a part of matrix $\frac{1}{p} R^2$ "covered" by R_y. (It should be noted that the index number (2) is the trace of matrix $R_y{}^{-1} Q_y$ itself.) In the special case when $Y = X$, this method leads to the theory of *component analysis*. This fact in itself seems to prove the adequateness of the definition (1).

By producing components y^* false interpretations can be avoided.

Results

To demonstrate the method a part experiment chosen from G. Láng's three-year (1964—65—66) fertilization experiments at Keszthely will be analysed. Trends of five characters in the wheat variety Fertődi 293 were studied under the influence of phosphorus fertilization. Treatments playing the role of the inducator variate were: absolute control and five further treatments: 0, 24, 48, 72, 96 kg/ha P_2O_5 + 50 kg/ha N + 62 kg/ha K_2O active agent. Characters examined (subject system X) : x_1 = ear number/m², x_2 = grain number/ear, x_3 = thousand-grain-weight, x_4 = hl-weight, x_5 = grain-straw-ratio. Three years averages of these characters are presented in Table 1.

Table 1

Three-years averages of characters

Number of treatment	x_1	x_2	x_3(g)	x_4(kg)	x_5
1	476.3	13.92	35.17	77.41	2.39
2	510.0	15.48	35.03	80.03	2.47
3	549.7	16.33	35.98	78.75	2.68
4	536.7	16.91	35.87	78.48	2.65
5	566.3	16.53	36.18	78.94	2.61
6	569.7	17.64	36.07	78.80	2.60

It was from this table that the correlation coefficients (r_{jk}) between variate pairs were computed; they are included in the matrix R, while the matrix $\frac{1}{5}R^2$ is given by percentage values.[1]

$$\text{R:}\qquad (r_{jk}) = \begin{pmatrix} 1 & .90 & .91 & .36 & .85 \\ .90 & 1 & .80 & .43 & .92 \\ .91 & .80 & 1 & .05 & .86 \\ .36 & .43 & .05 & 1 & .23 \\ .85 & .92 & .86 & .23 & 1 \end{pmatrix} \qquad {}^1\!/_5\,\text{R}^2\text{:}\qquad (q_{jk}) = \begin{pmatrix} 69.8 & 69.3 & 65.8 & 27.0 & 67.9 \\ 69.3 & 69.6 & 64.6 & 28.7 & 67.8 \\ 65.8 & 64.6 & 64.2 & 19.4 & 64.8 \\ 27.0 & 28.7 & 19.4 & 27.4 & 24.1 \\ 67.9 & 67.8 & 64.8 & 24.1 & 67.2 \end{pmatrix} \%$$

Figures falling in the main diagonal of the latter matrix directly show the information contained in each character on all five characters together. Accordingly, character x_1 contains the most information (69.8%), while x_4 the least (27.4%).

As to the joint information content of the two characters it follows from (2) as a special case ($r = 2$) that

$$I_{jk} = I(x_j, x_k \rightarrow X) = \frac{1}{1 - r_{jk}^2}(q_{jj} + q_{kk} - 2r_{jk}q_{jk})$$

---

[1] q_{jk} is the product the j and k lines of the matrix R, e.g.: $q_{23} = 100\,(0.90 \times 0.91 + 1 \times 0.80 + 0.80 \times 1 + 0.43 \times 0.05 + 0.92 \times 0.86)\% = 64{,}6\%$.

The calculations give us the following:

$$\begin{array}{llll} I_{12} = 77.2\%, & I_{13} = 82.9\%, & I_{14} = 89.3\%, & I_{15} = 77.7\% \\ & I_{23} = 84.6\%, & I_{24} = 88.7\%, & I_{25} = 78.4\% \\ & & I_{34} = 89.9\%, & I_{35} = 76.6\% \\ & & & I_{45} = 88.2\% \end{array}$$

When associating the variate x_4 with any other variate we obtain an informator containing nearly 90 per cent information. In such a case we can choose the character which it is the most justified to registrate precisely, or which is the least difficult to measure, to go with it.

The I_{234} information content of informator $Y = (x_2, x_3, x_4)$ shown "promising" by the matrix R was also computed, and the result obtained was 96.9 per cent. The missing 3 per cent is practically negligible.

Let us decompose the informator $Y = (x_3, x_4)$ to "disjunct" components.

In the case of a two-element informator the characteristic equation of matrix $R_y^{-1} Q_y$ can be directly stated, and its roots will give the information content of the components y_1^* and y_2^*, respectively. Namely, by making use of the fact that I_y is equal to the trace of this matrix, after some calculation we obtain:

$$det\,(R_y^{-1} Q_y - \lambda E) = \lambda^2 - I_y \lambda + \frac{det\, Q_y}{det\, R_y}\,.$$

In our case the characteristic equation is:

$$\lambda^2 - I_{34}\lambda + \frac{q_{33}\, q_{44} - q_{34}^2}{1 - r_{34}^2} = \lambda^2 - 0{,}8990\,\lambda + 0{,}1386 = 0.$$

Its roots are: $\lambda_1 = 0.701$, $\lambda_2 = 0.198$, that is $I\ (y_1^*) = 70.1\%$ and $I(y_2^*) = 19.8\%$ (when totalled they are really $89.9\% = I_{34}$).

Latent vectors are also simple to form. A brief calculation shows that the components of the latent vector belonging to λ_i are: $K_i(q_{34} - \lambda_i r_{34})$ and $-K_i\,(q_{33} - \lambda_i)$, where K_i is an arbitrary constant. To produce the components s_3 and s_4 the standard deviations of variates x_3 and x_4 are needed. Then the components are

$$y_i^* = K_i \left(\frac{q_{34} - \lambda_i\, r_{34}}{s_3}\, x_3 - \frac{q_{33} - \lambda_i}{s_4}\, x_4 \right) \quad (i = 1{,}2).$$

From Table 1 $s_3 = 0{,}49$, $s_4 = 0.84$, and thus:

$$y_1^* = x_3 + 0.22 x_4 \qquad\qquad y_2^* = x_3 - 1.41 x_4.$$

The constants K_1 and K_2 were chosen in such a way as to obtain 1 for the coefficient of x_3.

Fig. 1 shows the trend of y_1^* and y_2^*. The component y_1^*, which contains a 70 per cent information, shows well the — initially definite, then from Treatment 4 on moderate — increasing tendency of the characters of wheat as a response to the increasing supply of P_2O_5. The break occuring in Treatment 4 can be found in columns x_1, x_3, and x_4 of Table 1, too.

It is worth mentioning that the component including each of the five characters and thus containing maximum information (first principal component) contains only 4.4. per cent more information than y_1^*.

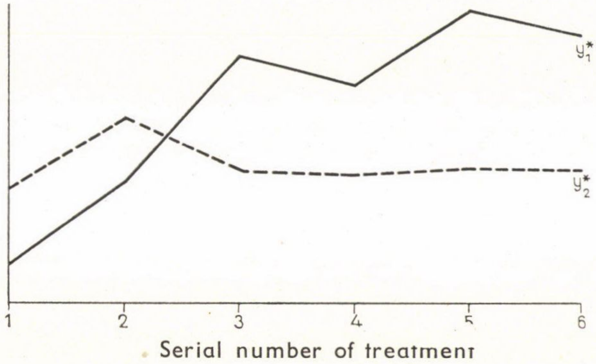

Fig. 1. Trend of components y_1^* and y_2^*.

The horizontal run of component y_2^* containing 20% information is broken only by Treatment 2. The same break is apparent with the hl-weight, and is present, though moderately, with the thousand-grain-weight. It can be said that y_2^* only gives information on the break occurring in the case of these two characters. The outstanding value of the hl-weight in Treatment 2 may further account for the result — unexpected for an agriculturist — that the hl-weight is present in most informative informators. Namely, this break is not indicated by any of the other characters.

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EFFECT OF CYTOSTATIC DIBROMOMANNITOL ON PROTEIN SYNTHESIS IN THE MYCELIUM OF BOTRYTIS CINEREA PERS. AND SCLEROTINIA TRIFOLIORUM ERIKSS.

By

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The authors have proved the influence of cytostatic dibromomannitol in increasing the level of protein nitrogen relative to the dry matter content by measuring the incorporation of radioactive carbon labelled glycine into proteins in mycelium cultures of two phytopathogenous micro fungi: *B. cinerea* Pers. *(Deuteromycetes)* and *S. trifoliorum* Erikss. *(Ascomycetes)*. It was characteristic that dibromomannitol — while in other tests inhibiting the synthesis of deoxy ribonucleic acid — increased the ratio of protein nitrogen in the dry matter content at all concentrations applied. This shift of more than 20 per cent in the proportion of the protein fraction is important from a theoretical aspect too.

Introduction

The cytostatic D-mannitol derivatives cause a decrease in the DNA, RNA and protein synthesis of animal and plant tissues *in vitro* in rabbit bone-marrow and tobacco callus cultures (Hidvégi—Lónai—Holland—Antoni—Institóris—Horváth 1967, Maróti 1967), and have — *in vivo* too — a growth inhibiting effect in higher plants (Péterfi—Brugovitzky—Kozma—Nagy Tóth 1959), as proved by Balogh—Frenyó (1967, 1970) who observed a decrease in catalase and polyphenol oxidase activities.

On the basis of their investigations Horváth—Institóris (1967) assume that in addition to their alkylating effect the dibromomannitol derivatives may exert a polarizing effect too by absorption through the lipoprotein membranes and nucleoproteids, and even cause wrong base coupling during the nucleic acid synthesis by changing the electron pattern of the macromolecules.

In our investigations we studied the effect of 1,6-dibromo-D-mannitol, or briefly dibromomannitol — a D-mannitol derivative of simple chemical structure — on the mycelium culture of two species of micro fungi. We wanted to find out how the development and protein synthesis of rapidly multiplying and growing heterotrophic lower plants (in the present case the mycelium cultures of two fungus species belonging to the *Ascomycetes* and *Deuteromycetes* respectively) change under the influence of this important cytostatic compound.

Material and Method

Two phytopathogenic micro fungi: *B. cinerea* Pers. (Deuteromycetes) and *S. trifoliorum* Erikss. (Ascomycetes) were used in our investigations. A culture medium free piece of mycelium, about the size of 1 mm^2, was taken from the stock culture and inoculated to the sterilized liquid culture medium (Treschow's synthetic basic culture medium modified at our Institute and standardized for micro and macro fungus cultures), in which dibromomannitol was dissolved at concentrations of 1, 10, 100, 1000 and 3000 ppm before sterilization.

The intensity of protein synthesis was followed by incorporating ^{14}C-glycin into the protein. The incorporation of labelled amino acid was determined after SCHMIDT—THANNHAUSER (1945) by fractioning proteins insoluble in 10 per cent TCA of +4 °C temperature, with modifications by PARTHIER (1961), OSBORNE (1962, 1965) and FLETCHER—OSBORNE (1965) taken into consideration. The specific activity of glycin-1-^{14}C was 26 mC/mM. The mycelium used for the test was floated for 4 hours on a solution of 100 ml volume and 0.5 μC/ml activity. The radioactivity of the fraction insoluble in trichloro acetic acid was determined with a Paccard-Tricarb instrument, by the method of liquid scintillation. Radioactivity was expressed in c.p.m./100 mg fresh weight.

Results

The dibromomannitol treatments exerted either stimulating or inhibiting effects depending on the concentration in cultures of both fungi examined *(B. cinerea* Pers. and *Sclerotinia trifoliorum* Erikss.). It must by all means be emphasized that a stimulation of the dry matter increase could be demonstrated even at the lowest concentration applied, while inhibition occurred only at the highest concentrations, at 1000 and 3000 ppm in the case of *B. cinerea* Pers., and only at 3000 ppm with *S. trifoliorum* Erikss.. Tables 1 and 2 clearly show the optimum effects exerted on dry matter increase, at 10 ppm with the *B. cinerea* Pers, and at 100 ppm in the case of *S. trifoliorum* Erikss. culture.

The intensity of protein synthesis measured with radioactive carbon incorporated was stimulated by all concentrations applied. The ratio between protein and non-protein nitrogen (NPN) increased characteristically and relative to the concentration, as did the percentage protein nitrogen in positive correlation with the total nitrogen in the case of *B. cinerea* Pers.

At the same time, with the other micro fungus examined *(S. trifoliorum* Erikss.), at optimum concentrations the ratio of TCA soluble and insoluble fractions indicated a considerable increase in the proportion of protein nitrogen.

The phenomenon is remarkable because in the case of higher animal and plant organisms the effect could be traced back directly to the inhibiting effect of dibromomannitol on DNA synthesis. The high biological effectivity can be tested with the inhibition of organic matter increase observed in the micro fungi examined too, nevertheless the unexpected stimulating effect of the lower concentrations applied on the proportion of total nitrogen to dry matter and of protein nitrogen within, leads to the possibility of drawing new conclusions.

Cytostatic effect as tested with organic matter increase and protein synthesis is very remarkable owing to the protein in the total nitrogen proportions.

Table 1

Effect of various concentrations of dibromomannitol on the dry weight increase of B. cinerea Pers. culture, and on the stimulation of radiocarbon labelled glycine accumulation and incorporation into the protein fraction insoluble in 10 per cent trichloro acetic acid, after 18 hours of exposition, expressed in 1000 cpm

Dibromo-mannitol ppm	Dry weight in g	Radioactivity in 1000 cpm				Percentage proportion of protein nitrogen related to total nitrogen
		TCA-insoluble fraction		TCA-soluble fraction		
		to total colony weight	in 100 mg	to total colony weight	in 100 mg	
0	0.265	175	66.04	69.7	26.30	71.52
1	0.380	280	73.68	112.0	29.47	71.43
10	0.431	625	145.00	173.8	40.32	78.24
100	0.375	540	144.00	136.1	36.30	79.87
1000	0.243	415	170.80	87.5	36.00	82.59
3000	0.108	312	288.90	64.2	59.44	82.94

With the activity values of radioactive isotopes standard deviation of the mean does not exceed 9 per cent.

It seems highly probable that in the case of dibromomannitol too, the unexpected increase of nitrogen metabolism observed is connected with DNA induction, as stated by van OVERBEEK (1966), nevertheless the study of the action mechanism requires investigations into the nucleic acid synthesis.

Table 2

Effect of various concentrations of dibromomannitol on the dry weight increase of S. trifolium Erikss. culture, and on the stimulation of radioactive carbon labelled glycine accumulation and incorporation into the 10 per cent trichloro acetic acid soluble nitrogen, and insoluble protein fraction after 18 hours of exposition, expressed in 1000 cpm

Dibromo-mannitol ppm	Dry weight in g	Radioactivity in 1000 cpm				Percentage proportion of protein nitrogen related to total nitrogen
		TCA-insoluble fraction		TCA-soluble fraction		
		to total colony weight	in 100 mg	to total colony weight	in 100 mg	
0	0.253	193	76.28	83.4	32.96	69.83
1	0.317	280	88.33	105.2	33.19	72.69
10	0.342	536	156.73	183.4	53.62	74.50
100	0.480	758	157.92	148.2	30.88	83.64
1000	0.410	613	149.51	114.5	27.93	84.26
3000	0.123	421	342.28	91.7	74.55	82.12

With the activity values of radioactive isotopes standard deviation of the mean does not exceed 9 per cent.

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COMPARATIVE ANALYSIS OF WHEAT STRAW BY THE METHOD OF QUANTITATIVE ANATOMY

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In the work here reported the Hungarian wheat variety Bánkúti 1201 and the Italian variety San Pastore were compared by the method of quantitative anatomy. Fibres were found to be longer in San Pastore. With the help of the strength-quotient (T-factor) developed by the author and his collaborator it was proved that San Pastore is more resistant to lodging. As the investigations were carried out on a very limited scale (on 2 specimens per variety) the obtained results are valid mainly for the plants examined. For general conclusions further investigations are needed. In the present report stress was laid on the description of the method itself. The anatomical method seems to be well-suited for expressing numerically the inclination to lodging in wheat and for the classification of the varieties as well. Variations in fibre length and cell wall volume are discussed.

Introduction

Wheat is and has been for a long time a factor of nation-wide importance in the cultivation of cereals in Hungary. It is thus understandable that much consideration has been given by both scientific and the professional circles to the problem of increasing the quantity and quality of wheat crops, and to the possibility of securing hardy varieties adapted for cultivation on different soils. Since the second half of the past century a great deal of work has been done in this field, leading to results not to be underestimated. Thus, e.g. it was in this country that the wheat varieties Székács and Bánkuti were produced, the latter of which deservedly acquired world-fame. Progress, however, must not stop and therefore much effort is being made to substitute Bánkuti by superior varieties, and there are two reasons to do so: the first is that Bánkuti is a tall-growing variety with relatively low yield, the second is that it inclines to lodging which is a strong drawback in machine harvesting. At present, the experiments are aimed at improving some new domestic varieties (e.g. Fertődi) with strong straw and at acclimatizing high-yielding foreign varieties (San Pastore, Besostaya). Wheat straw has recently gained industrial significance too, as in the factories for straw cellulose production mainly paper is produced. A better knowledge of the anatomical and quantitative anatomical characteristics of wheat straw is thus of industrial and agricultural interest. Now, part of the author's relevant investigations are reported in this paper.

In Hungary, Miltényi was the first to conduct methodical investigations on the anatomy of corn stalks (MILTÉNYI 1931). He studied stem development, activity of the intercalary meristem, distribution and the arrangement of the vascular bundles, formation of sclerenchyma, etc. Later on, wheat stalk was also studied by Mándy in connection with manuring experiments and he found the extent of manuring, the length and wall thickness of the fibres, and dimensions of the different tissues as related (MÁNDY 1954). At that time the author and his one-time collaborator, Gyula Pál, were engaged, also in the course of manuring experiments, in studying quantitative morphology and anatomy of rye straw (STIEBER—PÁL 1957). They noted an interesting parabolic relationship between the rate of elongation of the internodes in differently treated plants (1. c. p. 245—246), and an inverse relationship in the variations of internode length and fibre length. At the same time NILSON—JOHNSON—GARDNER (1957), working with several wheat varieties, reported on similar results with respect to cell length in the parenchyma, while KOLTAY—SIMON—PRÉCSÉNYI (1959), described, the variations in fibre length in reeds as originating from different habits. Then, again in relation with manuring experiments we studied the anatomy of wheat straw (STIEBER—PÁL 1959). It was found that, like in rye, cell wall volume and fibre length increased from the top of the stem downward, but decreased again in the lowermost internode (op. cit. p. 446; fc. STIEBER—PÁL 1957, Fig. 3. p. 248). These authors examined the mathematical correlation between lodging and anatomical structure in detail and constructed a mathematical formula allowing the calculation of the relative strength quotient (T) (op. cit. pp. 441—442). These results, when compared with those of Ulbricht's potassium fertilizing experiments (ULBRICHT 1937) were found to coincide on more points. Moreover, STIEBER—PÁL (1957, 1959) in both the rye and wheat examinations the sclerenchyma was found to form from the top downward an increasingly coherent ring which may even separate from the epidermis in the lowermost internode and a parenchymatous hypoderm may intercalate between them. Similar observations were reported on the stems of other *Gramineae* by BURDUJA—TOMA (1964, 1965). Later on a quantitative anatomical study was, made by HALÁSZ (1962) on wheat stem as a result of which he concluded that though the wall thickness of the fibres gradually increased towards the base, their length reached its peak in the medial internode. Subsequently, several wheat varieties were examined by SIMON-WOLCSÁNSZKY—CSÁSZÁR (1963), who obtained similar results with respect to fibre length and remarked in their report that this was the reason why they were not able to support the relevant findings of Stieber—Pál. It should be noted, however, that the latter authors referred to the inverse ratio between internode length and fibre length as to a general rule, all the more so as they characterized the curves for fibre length as expressedly asymmetrical maximum curves: "... these curves have to be regarded as asymmetrical maximum curves . . ." (STIEBER—PÁL 1959, 446). The curve obtained by SIMON-WOLCSÁNSZKY—CSÁSZÁR (1963, 411, Fig. 3) has the same shape, but both their curve 1, and the one obtained by HALÁSZ (1962, 202, Fig. 6), display a small peak above the lowermost internode. And if internode length is taken into account it will appear that the upper, i.e. third internode gets to the lower third or rather to the lower fourth of the stem (SIMON-WOLCSÁNSZKY—CSÁSZÁR 1963, 413). So even if fibre length is seen to reach its peak in this region, in reality the peak lies in the proximity of the stem base. A few years ago Haraszty and his collaborators (HARASZTY—SZABOLCS—OPPEL 1962, 1963; HARASZTY 1964) carried out fertilizing experiments on hemp (with fertilizers containing K-, N- and P) with regard to the dimensions and the distribution of the fibres. They reported that in the presence of sufficient quantities of N and P, potash fertilizers increased fibre dimensions. As regards the width of the fibres, Haraszty came to the same conclusions as Stieber and Pál, i.e. that cell wall volume increases downwards in the stem (HARASZTY 1964, 28), but according to Haraszty the increment of volume is continuous in direction of the plant base. A study on the histology of rice straw has been reported by SZEPES—HAJAS (1960). Now in Hungary Paál and Szabó conduct microscopical measurements on the leaf base and stem with the sclerenchyma belt in the cross section from the 3rd internodes of the wheat varieties Bezostaya 1 and Fertődi 293 (PAÁL—SZABÓ 1967).

Material and Method

In the present paper some quantitative histological features of the straw of the Hungarian wheat variety Bánkúti 1201 and of the Italian variety San Pastore are described. Both were grown side by side under identical conditions, on 3×10 m plots at the Agricultural Research Institute of the Hungarian Academy of Sciences, in 1960. From among the fully developed plants with mature spikes two specimens were pulled and their stems (A and B) examined. Therefore it should be emphasized that the results obtained permit only deductions as to the regularities in the histological features and that for conclusions of universal validity a larger number of specimens will have to be examined. Nevertheless the author considers it important to report on the results because it is through them that the methods of experimentation and evaluation can be illustrated.

From the internodes above ground of the specimens cross-sections and, using the Schultze-technique, macerates were made. From each of the longer internodes three preparations were made. One was removed from the geometrical center of the internode, a second and a third one from a distance of 1 cm from its lower and upper ends, respectively. From each of the shorter internodes only two preparations were made. They were removed from the boundary of the geometrical thirds of the internodes. In each macerate the length of 50 fibres was measured. A microscopical ocular integrator (SCHUCHARDT 1954) was employed for measuring the ratio of cell wall to cell cavity in the full cross-section surface of the sclerenchyma belt. The length and width of the internodes, the thickness of the sclerenchyma and of other tissue belts were also measured. The internodes were designated numerically from the top of the straw downward (n, n-1, n-2, etc.), while within the internodes the upper samples were marked with *f*, the middle ones with *k*, and the lower ones with *a*.

Results

Data of the examined plants:

	Bánkuti 1201		San Pastore	
	A	B	A	B
	cm		cm	
Plant height above-ground to spike base	77	76	67	57
Length of internode n	42.5	39	27.5	25
n—1	15.5	16	17	14
n—2	10	11	9	8
n—3	6.5	7	5	3
n—4	2.5	3	2	1
Level of above-ground sampling				
n/f	70	68	56	50
n/k	52	54	44	40
n/a	36	39	33	28
n—1/f	34	36	31	25
n—1/k	27	29	23	19
n—1/a	20	22	17	13
n—2/f	18	19	15	11
n—2/k	14	15	11	8

It appears from the Table that the two San Pastore plants differed in height by 10 cm. There were height differences of 3—5 cm at the corresponding sampling levels. The difference in height of the two Bánkuti plants was of 1 cm only and the differences in heights of the corresponding sampling levels did not exceed 2 cm. Therefore in the following comparison the histological numerical data for the two Bánkuti 1201 plants were, in some cases, reduced, whereas the values obtained for the San Pastore plants were treated separately.

Variations in fibre length as plotted against the internode levels are represented in Fig. 1. At n-1/a, viz. at level *a* of the second internode from above, all curves show significant maxima and for the Bánkuti plants *A* and *B* these are absolute maxima. The curves display a second maximum at n-3/a, viz. in the lower part of the second internode from the base upwards and for San Pastore B that is the absolute maximum San Pastore A reaches its absolute maximum in the lowermost internode, n-4. Thus the curves of Bánkuti display about the same shape as does the averaged curve No. 1. obtained by Simon-Wolcsánszky—Császár and the curve compiled by Halász. The curve

	Bánkuti 1201		San Pastore	
	A	B	A	B
	cm		cm	
Level of above-ground sampling				
n—2/a	10	11	8	5
n—3/f	8	9	6	3.5
n—3/a	3.5	4	3	1.5
n—4	2	2	1	0.5
Width at sampling level				
n/f	1.95	2.15	2.08	1.76
n/k	2.65	2.65	2.73	2.30
n/a	2	2.24	2.20	2.15
n—1/f	2.75	2.92	2.85	2.45
n—1/k	3.11	3.55	3.31	3.26
n—1/a	2·71	2.76	3.07	2.49
n—2/f	2.71	3.05	2.94	2.46
n—2/k	2.93	3.16	3.26	2.91
n—2/a	2.38	2.89	2.67	2.57
n—3/f	2.52	2.84	2.78	2.49
n—3/a	2.59	2.67	2.78	2.69
n—4	2.28	2.71	2.51	2.44

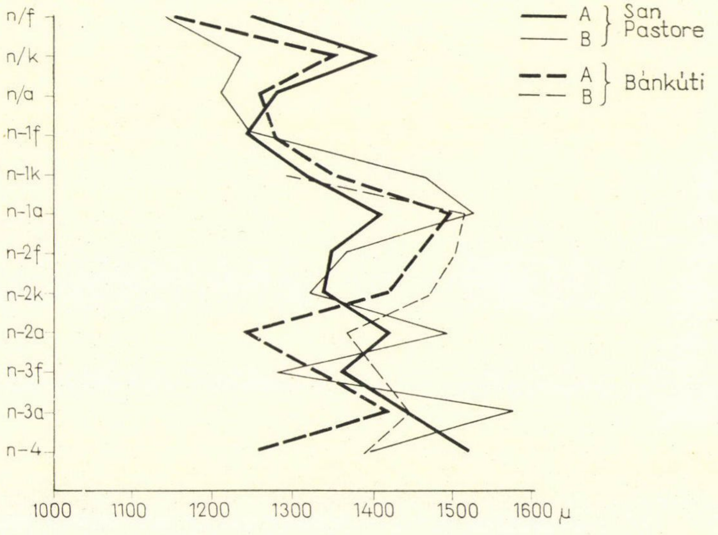

Fig. 1. Fibre length as plotted against internode levels n = upper internode, n-4 lowermost internode, f = upper, k = middle, a = lowermost internode level

of *San Pastore B* is similar to the difference, however, that in it, like in curve No. 2. Simon-Wolcsánszky it is the second, lower maximum which is greater. In other respects the curves of *San Pastore* are in good agreement with those obtained by STIEBER—PÁL (1959). Thus the suggestion that fibre length is inversely proportional to internode length is valid only for the San Pastore B and to a certain degree, for the San Pastore A plants, whereas the proposition that the longest fibres occur in the middle internodes is valid only for the two Bánkuti plants and even in their case with the modification that the maximum shifted upwards from the medial internode to internode n-1.

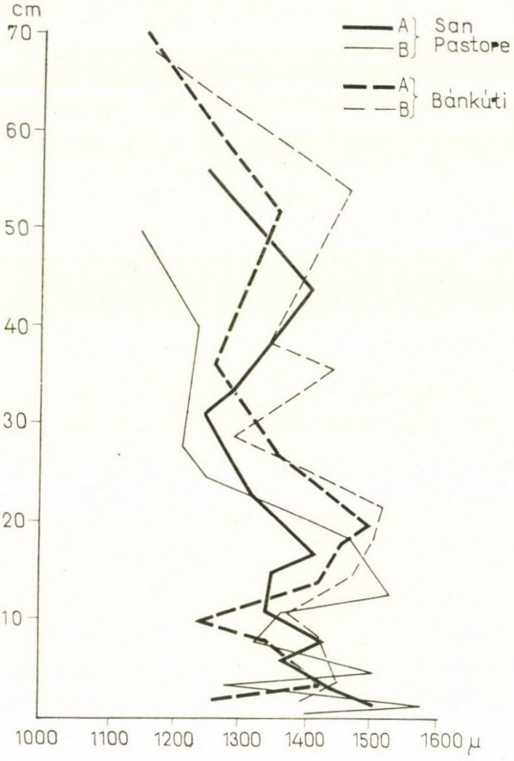

Fig. 2. Fibre length as plotted against aboveground height of the levels

If the values in the Table are now plotted against the aboveground height (Fig. 2) it will become evident that the relative and absolute maxima for the n-1/a levels occur in the curves of Bánkuti in the lower third and in those of San Pastore in the lower quarter of the straw, while the lower maxima (for San Pastore the absolute maxima) occur very near to the base, at 1—5 cm above the soil surface. Thus the proposition that fibre length increases in the successively lower internodes is valid for 70 per cent of the straw length in the Bánkuti, and for 96—100 per cent of the straw length in the San Pastore samples. However, fibres were found to be longest in the two San Pastore stems with values varying from 1500 to 1600 μ. In the Bánkuti stems these values hardly reached 1500 μ. The increment of length towards the base was quite substantial inasmuch as from a mean length of 1300—1400 μ the fibres increased to 1500—1600 μ.

Variations in cell wall volume in the sclerenchyma belt as plotted against the internode levels are represented in Fig. 3 and are seen to vary within a wide range since cell wall values of from 40 per cent to 80 per cent occur (see also Figs. 4, 5). The values increase towards the base and drop again in the lowermost internode. The highest maxima occur in internodes n-2 and n-3. A remarkable feature of the diagram is the consistently opposed shape of the curves for cell wall rate and fibre length (Fig. 6). Thus, e.g., cell wall rate in San Pastore reaches its maximum, while cell length drops to its minimum at the n-1/f level: at n-2/a cell wall rate is at its minimum, fibre length at its maximum, at n-3/f reversedly, etc. From the diagrammatic representation of the cell wall rate values plotted against the aboveground height (Fig. 7) it clearly appears that the rates increase with strong fluctuations consistently from the top downward along 90—95 per cent of the straw. In this respect the curve is in agreement with Halász' curve (Halász 1962, 203), except for the lowermost internode. In the cell wall rates of San Pastore and Bánkuti there are no essential differences. The graphs display more or less parallel

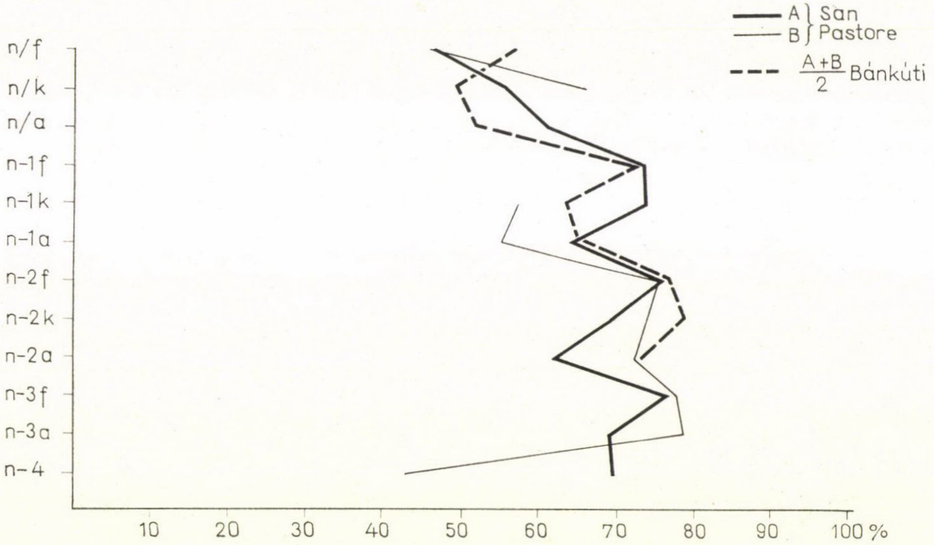

Fig. 3. Cell wall volume as plotted against internode levels. Key to signs in Fig. 1.

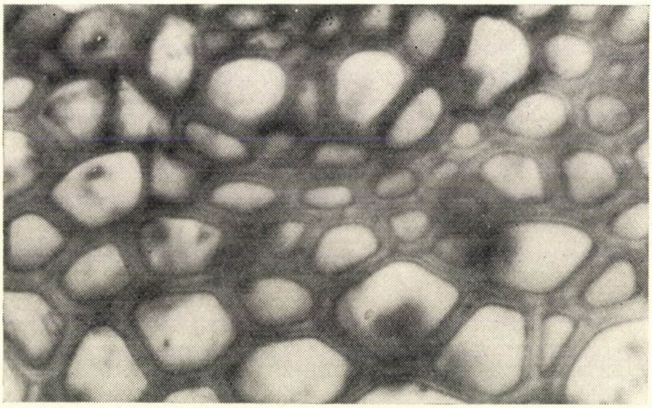

Fig. 4. Portion of sclerenchyma cross-section at internode level n/f in San Pastore wheat. Cell wall volume about 40 per cent. 1300 : 1

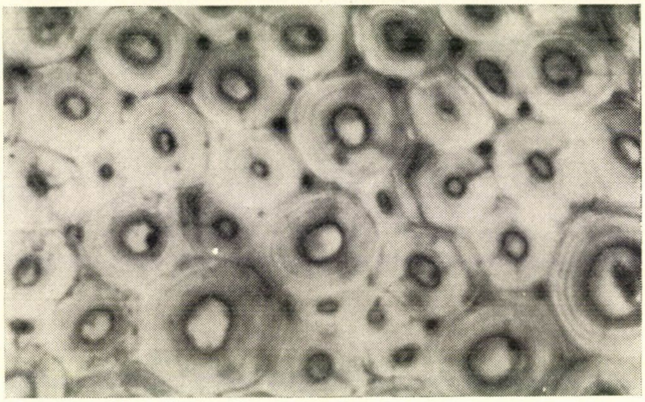

Fig. 5. Portion of sclerenchyma cross-section at internode level n-3/a in San Pastore wheat, cell wall volume about 80 per cent. 1300 : 1

trends; the absolute maximum appears, however, in the curve of Bánkuti B (it exceeds 80 per cent).

An attempt was made to evaluate the relative strength quotient (T) on the basis of a formula published in an earlier report on wheat anatomy (Stieber—Pál 1959, 441, formula No. 4). In the present work, however, multiplication with —1 in the numerator was omitted because when calculating with negative figures it would give results inconvenient for graphic representation. Instead, the base-line zero in the graph was changed to ±1. Thus the formula for the evaluation of T is:

$$T = \frac{N_h \times N_a^2}{N_s}$$

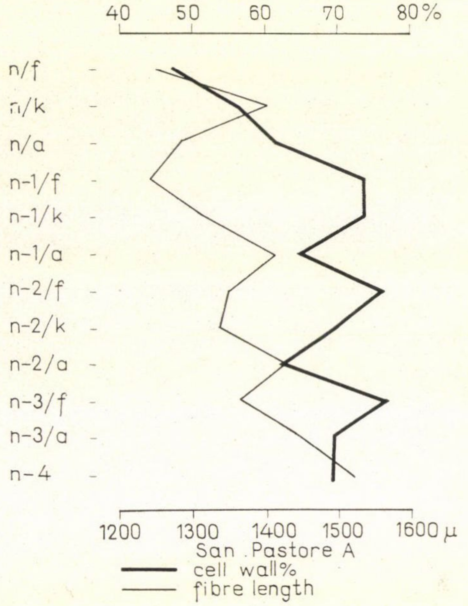

Fig. 6. Variations in fibre length and cell wall volume in stem of a San Pastore wheat sample
Heavy line: cell wall volume (%), light line: fibre length (μ)

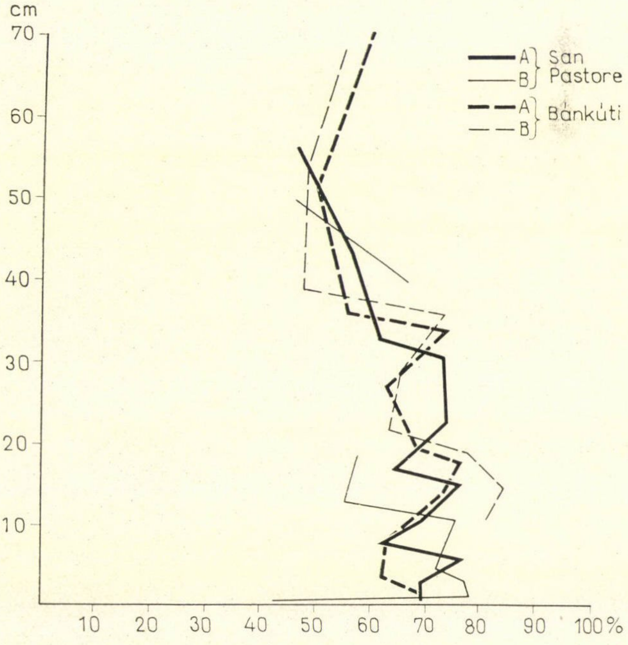

Fig. 7. Cell wall volume as plotted against above-ground height of the levels

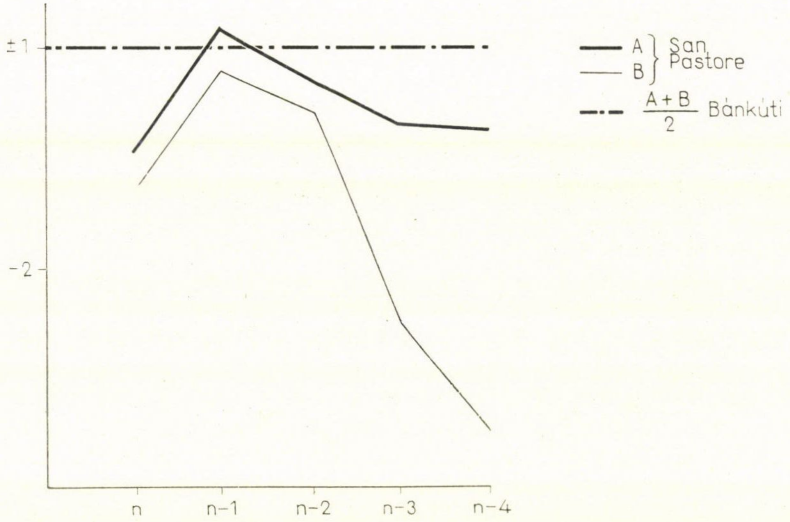

Fig. 8. Internode length quotients (N_h). Key to signs in Fig. 1

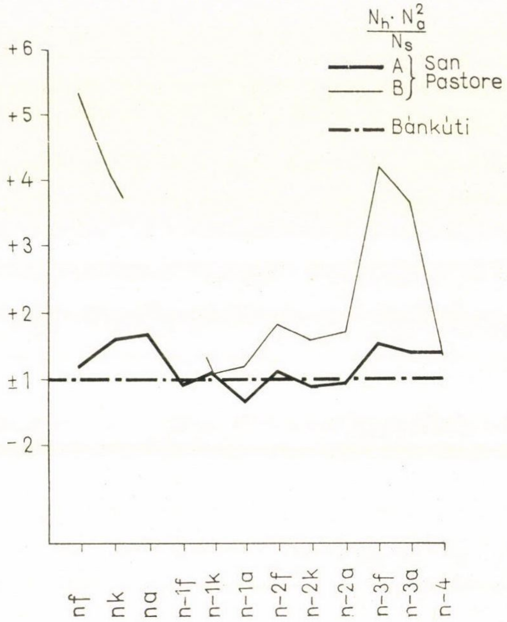

Fig. 9. Relative strength quotient (T). Key to signs in Fig. 1

where N_h stands for the internode length quotient, N_a, for the internode diameter quotient and N_s, for the cell wall volume quotient. The problems arising in connection with the calculation of these partial quotients and of the T-quotient will be treated in a subsequent report. The N_h-quotients, however, are represented diagramatically, in the present paper (Fig. 8). In contrast to the parabolic curves obtained for rye (STIEBER—PÁL 1957, 246, Fig. 1) and for wheat (STIEBER—PÁL 1959, 432, Fig. 1) the curves in this diagram are maximum curves lying in the negative range displaying their maxima precisely at the internodes n-1. It should be noted as a point of interest that the curves for fibre length also have their maxima at the same level (Fig. 1).

Curves T are shown in Fig. 9. It is noteworthy that both curves for San Pastore are minimum curves in the positive range, with one of their rising branches in the proximity of the straw base and the other in that of the straw top, at heights of 3—4 units and that only the minimum of the plant A comes to lie lightly (by a few tenths in the negative range. Although curve B displays a significant drop at internode n-1, the curve itself lies in the positive range.

Discussion

The question of regularity of the variations in fibre length along the straw is still open to discussion. In spite of the fact that not all of their diagrams for rye did follow the same pattern, STIEBER—PÁL (1957, 248, Fig. 3) stated that as a rule, internode length and fibre length vary in inversely. In a subsequent study (STIEBER—PÁL 1959) the authors made it clear that such a relationship is illustrated by one part of asymmetrical maximum curves only. Halász—Simon-Wolcsánszky have obtained maximum curves, but each of these curves displays a lower maximum, too, and a further curve compiled by Simon-Wolcsánszky is asymmetrical and shows a lower maximum. In the present report some curves for fibre length show middle or lower maxima, some show a positive gradation downward, so that two or even three types of curves may occur. In author's opinion all of them are resultant curves one factor of which is the internode length (as the growth factor), while the other is for the time being unknown. Depending on the magnitude of these two components the maximum can shift upwards or downwards to the base. This, of course, is only an assumption. It is a different elucidation of the problem when fibre length is considered in relationship to aboveground height. In that case cell length increases from the top downwards along the whole length of the straw or at least along the upper two- third or three-quarters of the straw, decreasing only in its lower third or fourth or, in some cases, at an even lower level. This regularity was clearly demonstrated by all results obtained in this country (STIEBER—PÁL 1957, 1959, HALÁSZ 1962, SIMON-WOLCSÁNSZKY—CSÁSZÁR 1963).

The shapes of the graphs for cell wall volume vary in general similarly to those for fibre length. Their sections, however, display a shape significantly and consistently opposed to those of the curves for fibre length. Although these data are too meagre to permit generalisations, still, it is worth taking notice of this phenomenon which points towards a possible relationship with the growth capacity of fibres. In the longer fibres the greater part of the capacity is supposed to get consumed to increase the length, thus in the shorter ones more capacity would remain available for increasing the volume of the cell wall.

The relative strength quotient (T-factor) indicates that San Pastore is by far less prone to lodging than Bánkuti 1201 (as shown by the examined specimens). This conclusion is substantiated by the position of the curve T in the positive range and by the high T-values for the lower internodes. Fibres and turgor are the two components which determine strength in wheat straw. In the earlier stages of development it is the turgor component, in the later stages the fibre component, which is greater and in the last stage the turgor component drops to zero, and this is the critical period when in case of averse weather conditions wheat may irreversibly lodge. If, on the other hand, weather conditions are favourable even wheats otherwise more inclined to lodging remain erect. Therefore with the aid of simple management experiments, even if they should be extended over a number of years, it may not be possible to determine whether a wheat variety is prone to lodging or whether the introduction of certain agricultural methods would increase or decrease this property. The author suggests that the application of the T-factor is more expedient than the former methods and allows in case of appropriate sampling and of a sufficient number of specimens safe conclusions to be drawn: the more so as the T-values being suitable for expressing relative strength numerically, varieties or even cultural techniques can be numerically classified in this way. Although the T-formula can be used also in its present form its further development, checking and improvement would seem advisable, all problems with which the author himself is also concerned.

Summing up what has been reported in the foregoing, it should be emphasized that the above results are suitable, first of all, for an illustration of the quantitative anatomical method but that owing to the limited number of samples less suitable for generalizations.

Acknowledgements

The author wishes to express his appreciation to Mr. B. Penke, his one-time pupil and to Miss I. Molnár for assistance in this work. Grateful thanks are also due to Mr. S. Rajki, Director, Agricultural Research Institute of the Hungarian Academy of Sciences, Martonvásár, for supplying the material utilised in this investigation and to Mr. S. Sárkány, director of the Institute for Applied Botany and Histogenesis, University Eötvös Loránd, Budapest.

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POLLEN TUBE FORMATION IN PEARS

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In 1968 and 1969 the relation of pollen tube formation to saccharose concentration, time of tube formation and temperature was studied in the pear varieties "Clapp kedveltje" and "Vilmos". Both varieties showed the maximum tube formation percentage with a saccharose concentration of 15 per cent. A linear correlation was found between percentage pollen tube formation and time. Pollen tube formation attained a maximum in 120 and 160 minutes respectively. Correlation between the rate of pollen tube formation and temperature can be illustrated by an optimum curve. The optimal temperature of tube formation was 23 °C in the variety "Clapp kedveltje", and 25 °C in "Vilmos".

Introduction

Results of pollen physiological studies performed with various fruit species and -varieties under different ecological conditions and with different methods are highly varied (NYÉKI 1970, 1972). Correlations between temperature and pollen tube formation as well as between the sugar concentration of the culture medium and the dynamics of tube formation are not clarified in the fruit species and -varieties. The above difficulties made the methodological studies concerning the pear varieties necessary.

Material and Method

The examinations were carried out with the pollen of the diploid pear varieties "Clapp kedveltje" and "Vilmos". The pollen was collected from five trees of each variety of a variety collection grafted to wild pear seedling stocks planted in 1953 at the Érd-Elvira experiment station of the Horticultural Research Institute. Some 5—7 days before flowering bearing shoots were taken from all cardinal points of the middle zone of the crown and brought to flowering under laboratory conditions at 20 °C. Average pollen samples were taken from the bearing spurs, from the lateral flowers of the third storey of the cluster.

Relation of pollen germination to time was observed with VISSER's (1955) method. The pollen was placed in drops and watched until tube formation started. From that time tube development was registered and proportions computed every five minutes. Three drops were examined in each variety, the rates of tube development were determined on the basis of 2×50 pollens per drop. Examinations were carried out in a thermostat, at a temperature of 23—25 °C. For the purpose of establishing the relationship between the rate of pollen tube formation and temperature the pollen was kept for 4 hours in a 15 per cent solution of saccharose at temperatures of 5, 10, 15, 20, 23, 25 and 30 °C until counted. The relationship between tube formation and saccharose concentration was studied in saccharose solutions of 0, 5, 10, 15, 20, 25 and 30 per cent respectively, at 23—25 °C. Pollen tube formation was determined after 4 hours; under optimal conditions this time was sufficiently long for tube development to attain a maximum.

Results

1. *Relationship between the rate of pollen tube formation and the concentration of saccharose.* The authors found different sugar concentration optima in the fruit species (FLORIN 1923, KOBEL 1926, 1927, ZIEGLER—BRANSCHEIDT 1927, KAMLAH 1928, MOFFET 1934, GRIGGS *et al.* 1953, REMY 1953, STÖSSER 1966, PEJKIC 1968, 1969, etc.). The optimum concentration of saccharose found by the above authors ranged between 10 and 15 per cent in most fruit species.

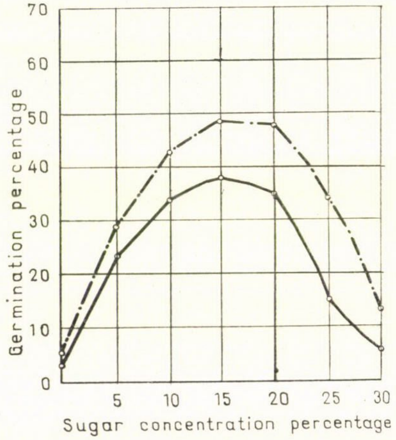

Fig. 1. Relationship between the rate of pollen tube development and concentration of saccharose at a temperature of 23—25 °C after 4 hours of tube development in 1968. (——— Clapp, —.—.— Vilmos)

According to our observations both diploid pear varieties examined attained the highest rate of tube development at saccharose concentrations of 10, 15 and 20 per cent (Fig. 1). These data are in unison with the saccharose concentration values found by DURAKOVICS (1957), NAGY (1960) and CHOLLET (1965) in various pear varieties.

2. *Dynamics of pollen tube development rates.* The length of time after which the rate of pollen tube development is determined is very important from the point of view of evaluation. REMY (1953) kept the pollen of various stone fruits in sugar solution for 6 hours before beginning examinations, VISSER (1955) started the examinations on the pollen of pear after 3—4 hours. According to their investigations tube development rates attained a maximum practically in 120 and 160 minutes, respectively, in both varieties (Fig. 2). Results show a linear correlation between the rates of pollen tube development and time — within the mentioned intervals of 120 and 160 minutes.

3. *Relationship between the rates of pollen tube formation and temperature.* According to the data in the literature the temperatures at which the authors

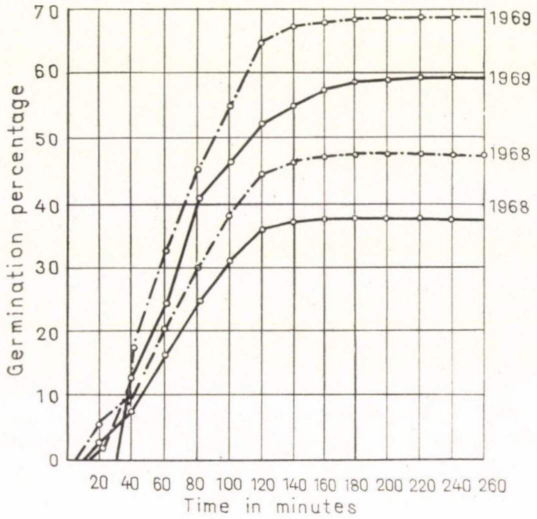

Fig. 2. Dynamics of the rate of pollen tube development in 1968 and 1969, at a saccharose concentration of 15 per cent and temperature of 23—25 °C. (——— Clapp, —.—.— Vilmos)

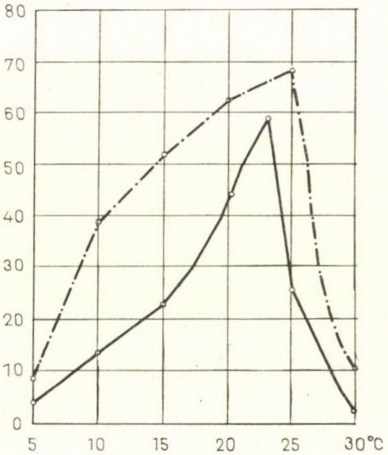

Fig. 3. Relationship between the rate of pollen tube development and temperature in 1968 and 1969, at a saccharose concentration of 15 per cent, after 4 hours of tube development. (——— Clapp, —.—.— Vilmos)

obtained maximum rates of pollen tube development varied from species to species. Remy (1953) performed pollen germination with stone fruits at 22—23 °C, Visser (1955) with pears at 23 °C, Durakovics (1957) at 21 °C and Nagy (1960) at 18—21 °C. According to our investigations (Fig. 3) optimal pollen tube formation in pears as related to temperature proved to be a specific character of the variety. The optimal temperature of pollen tube formation was 23 °C in the variety "Clapp kedveltje", and 25 °C in the variety "Vilmos". The correlation found between the rate of pollen tube formation and temperature can be illustrated with an optimum curve. The correlations correspond to VISSER's (1955) observations.

Conclusions

Saccharose concentrations of 10, 15 and 20 per cent are equally suitable for the purpose of pollen study in pear varieties. There is a linear correlation between the rate of pollen tube formation and time. Tube development rates of the varieties practically attain a maximum in 120 and 160 minutes. Correlation found between the rate of pollen tube development and temperature — which proved to be a specific character of the variety — can be illustrated with an optimum curve.

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A NEW ARTIFICIAL HYBRID OF SPECIES FROM THE GENERA FESTUCA AND LOLIUM (FESTUCA PRATENSIS HUDS. × LOLIUM TEMULENTUM L.)

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In the years 1969 and 1970 species of the two genera were crossed in various combinations. After the hand castration and isolated pollination of the flowers, by part of the hybrid embryos raised in vitro, hybrids were produced in several variations, one of which proved especially valuable. This intergeneric form was obtained by *F. pratensis* (2×) and *L. temulentum* (2×) crossed. Observations related to the hybrids are presented in the paper.

Introduction

Certain species of the genera *Lolium* and *Festuca* are readily crossed in situ too. Forms known as *Festuca loliacea* Curt., *Festucololium ascendens* (Reh.) A. et G. etc. in the literature were described by the botanists as early as around the turn of the century.

Since the fundamental works of MC ALPINE (1898, In: HERTZSCH 1959), ASCHERSON—GRAEBNER (1898—1902), JENKIN (1924, 1933, 1935), KNOLL (1928), HERTZSCH (1938), BULASEVIC (1938), WINKLER (1938) numerous interspecific and intergeneric hybrids have been produced by researchers. According to the authors' earlier summarizing work (HESZKY 1971) intergeneric hybrids have been successfully produced so far in forty four combinations. There are, however, only fifteen variations among them in which one or the other ploidy level form of *F. pratensis* occurs.

F. pratensis was successfully crossed with the species *L. perenne*, *L. multiflorum*, *L. persicum*, *L. oldenburgicum* (ESSAD 1956, 1960, WIT 1959, 1964, SULINOWSKY 1956, 1967, etc.), when used as pollen parent, and with various ploidy level forms of *L. perenne*, *L. multiflorum* (JENKIN 1955a, HERTZSCH 1959, 1960) when used as seed parent.

The crossing of *L. temulentum* L. — unlike the better known *Lolium* species — was not successful either with *F. pratensis* or with the other *Festuca* species.

According to JENKIN (1935, 1955b, 1959), in the case of *F. rubra* and *F. partensis* pollinated with *L. temulentum*, no development of either the embryo or the endosperm could be observed. In the variation of *L. temulentum* × *F. pratensis* the development of caryopses and embryos could be observed, however, the developed embryos were unviable.

Material and Method

From the species *F. pratensis* the Hungarian diploid ($2n = 2\times = 14$) variety "Gruber", while from the *L. temulentum* a diploid ($2n = 2\times = 14$) ecotype were used in our crossings.

At the time of flowering the transplanted individuals of the seed species were isolated with cellophane bags, in a greenhouse. The isolated flowers had been castrated by means of pincers, then — when flowering — pollinated according to CARLBOM's (1968) method. When fully mature the seeds were removed from the plants. The caryopses were placed in sterile Petri-dishes, on filter paper soaked with water, and the seedlings grown from the germinated seeds planted into the soil (HESZKY 1971).

Stainability of the pollen was studied in a preparation stained with carmine acetic acid.

The length of the stoma was measured with a Zeiss 15× micrometer ocular, in preparations made of the epidermis of the first leaf below the inflorescence, collected from shoots at the beginning of flowering.

Results

In 1969, 160 (100 per cent) flowers of *F. pratensis* were castrated and pollinated with the pollen of *L. temulentum*. After pollination signs of fertilization were found in 17 flowers (10.6 per cent), however, when mature, only 5 caryopses (3 per cent) contained embryos and a little endosperm. When germinated in the spring of 1970 two grains (1.2 per cent) proved viable, but a fully developed plant could be raised from only one of the seedlings (0.6 per cent).

The morphological features and physiological characters of the two parent species were very different, so the characteristics were expected to appear in highly diversified forms in the hybrid.

In the first year *F. pratensis* developed no flowering stalk, while both *L. temulentum* and the hybrid flowered already in the first year. It is probable that the hybrid — like the pollen parent and unlike *F. pratensis* — requires higher temperatures in the heat phase.

L. temulentum — in accordance with its therophyte character — withered after flowering, while the hybrid did not do so, but — after having been cut — shooted again. Thus the hybrid — like *F. pratensis* — has a longer lifetime.

A new characteristic different from that of the two parent species was observed in the hybrid, namely, the latter flowered in the second growth and — when cut repeatedly during the year — always developed a flowering after growth. In this characteristic the hybrid differed from both parents, since *F. pratensis* developed only leaves and *L. temulentum* withered after having flowered and been cut.

Fig. 1 shows that the stalk of the hybrid — like that of *L. temulentum* — is stiff and upright, but longer than that of either parent. Its tuft formation capacity shows a transition between the two parents. Its leaves are dark green as in the pollen parent, but broader and thicker than those of either parent.

The length of the stomata in the leaf epidermis is similar to that in *F. pratensis*. On the average of a hundred measurement data the stoma length was 46.5 micron (37.5 — 56.25 micron) with *F. pratensis*, 49.75 micron (43.75 — 58.75 micron) with the hybrid and 64.25 micron (54.5 — 75.0 micron) in the case of *L. temulentum*.

The inflorescence of the hybrid form a poorly branching cluster of a size exceeding that in both parents (Fig. 2).

Fig. 1. F. pratensis (2×) × *L. temulentum* (2×) hybrid and the parent species. From left to right: *F. pratensis* (when flowering), hybrid (when earing), *L. temulentum* (when flowering)

Fig. 3 shows that the spikelets of the hybrid are longer than those of both parents and — unlike *F. pratensis* — awned, but the length of awns does not attain that of *L. temulentum*. The bract scales are of intermediate size between the two parents, but — unlike in the parent species — reach to the middle of the spikelet. The number of flowers in the spikelets of the hybrid is the highest of all. The shape and size of the foral glume are similar to those in *L. temulentum*, but at the time of the dehiscence of the anther open in the same way as in *F. pratensis*.

The pollen grains of the hybrid are 100 per cent sterile. Back-crossing to the parent species was useless, so there has been no possibility to study the F_2 and F_3 generations.

Fig. 2. Inflorescence of the *F. pratensis* (2×) × *L. temulentum* (2×) hybrid and the two parent species. From left to right: *F. pratensis*, hybrid, *L. temulentum*

Fig. 3. Spikelet of the *F. pratensis* (2×) × *L. temulentum* (2×) hybrid and the two parent species. From left to right: *F. pratensis*, hybrid, *L. temulentum*

When summing up the results of our observations we can characterize the phenotype of the *F. pratensis* (2×) × *L. temulentum* (2×) hybrid as follows:

1. *Features characteristic of F. pratensis:* size of epidermal stomata.

2. *Features characteristic of L. temulentum:* higher temperature requirement in the heatphase, colour of leaves, stiff and upright character of stalks (Fig. 1), awned spikelets (Fig. 3), shape and size of foral glume (Fig. 3).

3. *Intermediary features:* length of leaves (Fig. 1), length of awn (Fig. 3), opening of foral glume at the time of flowering.

4. *Features exceeding by order of magnitude those of the two parents:* length of stalk (Fig. 1), length of inflorescence (Fig. 2), length of spikelet (Fig. 3), width and thickness of leaves (Fig. 1), number of flowers per spikelet (Fig. 3).

5. *Feature appearing in the hybrid and not characteristic of either parent:* regeneration of a flowering aftergrowth.

The hybrid possesses a number of economically valuable characteristics too, therefore the amphiploid form should be produced in the future in order to improve fertility.

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IMPORTANCE OF INTERACTION IN IMPROVING THE PROTEIN CONTENTS OF MUTANT POPULATIONS

By

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The low protein content "Horpácsi kétsoros" winter barley variety was exposed to EMS treatment in order to induce genotypic variation in the protein content. Protein examinations were carried out in the M_2 plants of 90—100 percent ear fertility and their M_3 progeny lines. The average protein content of M_2 and M_3 populations was 29 percent higher than that of the low protein content control (11.2 percent). In the genetically heterogenous mutant population the variance of interaction between genotype and year was found to be remarkably high compared to the variances of genotype and error. In this population, therefore, only a moderate progress can be expected of selection. After the mutant population had been separated into populations of low- and original genotype × year interaction, the average protein contents of the two populations remained perfectly identical, but the predicted genetic progress improved to a very high extent in the low interaction population. Efficient selection thus requires not only a favourable line average but also a low interaction between genotype and year as preconditions.

Introduction

"Horpácsi kétsoros" (double-row), a winter barley variety with strong straw, good productivity and phenotypic stability, but low protein content, was treated with EMS in order to induce genotypic variation in the protein content, thus creating a genetic basis for improvements in the protein content. The possibility of increasing the protein content has been referred to by many authors. In Hungary POLLHAMER (1970) presented results concerning the protein content of elite plants selected from the X_2 generation of four winter barley varieties treated by X-ray. Induced micro-mutant strains contained a maximum of 16—18.5 per cent protein.

Among the cereals the protein mutation induced by gamma irradiation in the "Sonora 64" two-gene-dwarf wheat variety should be mentioned here the average protein content of which was increased by SWAMINATHAN (1968) from 14.5 to 16.5 per cent.

The endospermic protein fractions of the M_3 population of wheat grains treated with EMS were found by BHATIA *et al.* (1970) to be of a higher variability than those of the control.

Material and Method

5000 grains of the winter barley variety "Horpácsi kétsoros" were placed in a 0.1 per cent EMS solution and — while constantly shaken — treated for 24 hours at 20 °C. The M_2 and M_3 generations were sown in the autumn of 1969 and 1970 at Kiszombor, at a spacing of 40 cm between rows and 10 cm in the rows. In 1970 fertility of the main ear in the M_2 population was determined by the ratio of flowers and grains set. Crude protein examinations were carried out with M_2 plants of 90—100 per cent fertility and their M_3 progeny lines, in two replications each, after Kjeldahl's method. Crude protein data apply to a 100 per cent dry matter content.

Genotypic variance of lines, variance of genotype x year interaction, and error variance were determined on the basis of the following model (Table 1).

Table 1

Model of variance analysis

Source of variance	Degree of freedom (d. f.)	Variance components
Lines	(V – 1)	$\sigma_e^2 + R\,\sigma_{gy}^2 + RY\sigma_g^2$
Lines × Year	(V – 1) (Y – 1)	$\sigma^2 + R\sigma_{gy}^2$
Error	(R – 1) (VY – 1)	σ_e^2

V = number of lines; R = number of replications; Y = number of years.

Phenotypic variance of line-averages was determined by the following formula, after Rasmusson—Glass (1967): $\sigma_{ph}^2 = \sigma_g^2 + \frac{\sigma_{gy}^2}{Y} + \frac{\sigma_{e}^2,}{RY}$ where σ_g^2 = genotypic variance, σ_{gy}^2 = variance of line x year interaction; σ_e^2 = error variance.
$H = \sigma_g^2/\sigma_{ph}^2$. Predicted genetic progress $(G_s) = Hk\sigma_{ph}$, where $k\sigma_{ph}$ is the selection differential expressed by phenotypic variance.

The data examined were of normal distribution.

The total and partial sterility induced by EMS inhibited efficient selection in the M_2 generation. According to our investigations 26.8 per cent of the M_2 population could be placed in the 90—100 per cent fertility interval, and these plants exceeded the control by 11.2 per cent with the grain yield of their main ears, while populations of lower fertility were much inferior to it. Ear productivity is an important yield component, therefore, from the point of view of an accelerated genetic progress, M_2 plants of 90—100 per cent fertility and their progeny lines were considered to be a basic material suitable in which to study the genotypic variation of crude protein.

The possibility of improving protein content in the winter barley variety "Horpácsi kétsoros" by EMS treatment was studied with the M_3 and M_4 grains of the M_2 and M_3 generations (Szirtes 1971). In the genetically heterogenous mutant population the variance of genotype x year interaction was remarkably high compared to the variance of genotype and error. In the mutant population interaction variance was 5.1-times higher than error variance. With a steadier phenotypic manifestation of protein content in view, selection of genotypes in which the high protein content changes but slightly from year to year is an important breeding task.

The extent of genetic progress will be studied subsequently in lines with moderate genotype x year interactions.

Results

In lines with low interaction between genotype and year average crude protein content in the M_3 and M_4 grains of the M_2 plants and M_3 lines was 14.45 per cent (Table 2). Protein content in this mutant population is 29 per cent higher than the 11.2 percent crude protein content of the initial winter barley variety "Horpácsi kétsoros".

Table 2

Variance components of crude protein content and values of expected genetic progress

Designation	Number of lines	Number of protein tests	$\overline{X}$	s^2_g	s^2_{gy}	s_e^2	s^2_g/s^2_{ph}	G_s	Expected average of progenies	Percent of G_s to average
Horpácsi kétsoros (controll)	10	40	11,20	0	0.352	0.437	—	0	11.20	—
M_2 plants and M_3 lines	116	464	14,43	0.115***	1.404***	0.276	13	0.25	14.68	1.7
M_2 plants and M_3 lines (low interaction of genotype x year)	50	200	14.45	0.964***	0.088	0.235	90	1.62	16.07	11.2

*** P = 0,1%

A considerable extent of the variance in lines is mostly of genotypic origin. 90 per cent of phenotypic variance consists of genotypic variance. In the case of a 10 per cent selection intensity the estimated genetical progress is 1.62 g crude protein per 100 g dry matter, which is 11.2 per cent of the average crude protein content. 16.07 percent crude protein is expected to be contained in the progenies selected.

Conclusions

Under the influence of the EMS treatment applied to the low crude protein content winter barley variety "Horpácsi kétsoros" average crude protein content in the M_2 and M_3 mutant populations increased considerably (by 29 per cent) compared to the control. Apart from this, further genetic improvement may be attained by exploiting the genotypic variation brought about by mutagen treatment. According to our investigations, when selection—

on the basis of two-year average protein contents in lines — was made in mutant populations in which lines with low and high genotype × year (g × y) interactions occurred, then selection was expected to result in a low extent improvement of average protein content in the progenies.

When the mutant population was separated into population of low- and original g × y interaction then — while the average crude protein content remained totally unchanged in both populations — the efficiency of selection greatly improved in the population of low g×y interaction.

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A NEW METHOD OF STUDYING THE SWARMING OF EPICOMETIS HIRTA PODA

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During 1970 and 1971 a new forecasting method was developed for studying the swarming dynamics of *E. hirta* imagos: the blue water-dish colour trap. In the course of investigations blue colour — similar to the colour of the cornflower — proved to attract the insect pest with success. White, yellow, green and red colours were found to have no attraction. The blue dish successfully collected various *Hymenoptera* species occurring in the orchard too. Trapping is considerably influenced by microecological conditions, therefore the study of *E. hirta* infestation in a plant stand requires the application of more than one trap.

Introduction

E. hirta attacks various cultivated plants (rye, fruits, ornamentals, etc.) causing damages varying from year to year (Martinovich 1962). The introduction of efficient control methods and studies on the economical aspects of control require adequate methods of forecasting and the investigation of population dynamics. No information on such methods has been found in the literature. In Hungary Joachim—Josepovits (1951) studied the colour sensitivity of the insect, however, it proved indifferent in the tests. When studying the colour sensitivity of the insects Mazohin—Porsanjakov (1971) found that they could distinguish the blue colour from the others well. The colour sensitivity of insects and the attractiveness of colours have been utilized by many in collecting insects and studying swarming (Chauvin 1967, Jorgensen 1962, Moericke 1950, Schreier 1963, Southwood 1966, etc.). Most of them were concerned with the effect of yellow light-traps especially suitable for collecting aphids and bugs. A blue trap was used for the collection of frit-fly by Southwood sen. (1966).

Material and Method

In the colour-trap studies six cornflower-blue plastic dishes of various depths were used; one of them was 30 cm, the other five 12 cm diameter. The dishes were filled with drinking water and placed on a wooden stand at a height of 60 cm above ground (Fig. 1). The colour-traps were operated in 1970 in a mixed orchard among peach-trees, in 1971 partly in the same place, partly at the edge of a homeplot vineyard among currant-bushes, at a distance of 2 m from one another, with plum- and peach-trees nearby. Beside the blue colour attraction 24 yellow, white, red and green light-traps of various shape and size was also studied. The insects trapped were counted and removed from the water of the traps every day (Fig. 2).

Fig. 1. Trapping dishes of various colour placed among peach-trees

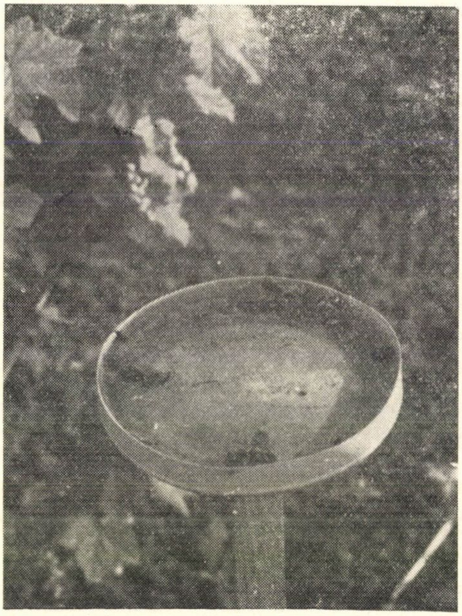

Fig. 2. Result of one-day trapping in the dish

Results

During investigations in 1970 and 1971 the use of the blue water dish gave useful information on the course of swarming of *E. hirta* imagos. By means of these traps 127 *E. hirta* imagos were collected during the two years of the investigations. A complete observation was only made in 1971, when swarming was successfully followed from the very beginning to the end (Fig. 3). Swarming began on 10th April, the peak was shown between 15th and 20th April. From then on some swarming was still observed till the end of May. The results of traps placed at a distance of 2 m from each other highly fluctuated (Table 1) probably due to microecological causes. Beside the blue

Table 1

Trapping by five blue light-traps of identical colour and size

Traps:	1.	2.	3.	4.	5.
Total number of trapped imagos	6	5	13	34	19
Average trapping in ten days	1.1	1.0	2.6	6.8	3.8
Maxima and minima of trapping per day	0—2	0—2	0—4	0—5	0—4

light-traps the attraction of red, green, white and yellow colours too was studied; as opposed to the 127 specimens trapped by the blue colour, only one *E. hirta* imago flew into a trap of different colour.

Simultaneously with the study 22 imagos were collected with a blue drinking glass placed under a peach-tree. While studying the swarming, further data were obtained as to *E. hirta* flying almost exclusively in the daytime (around noon). Swarming is often interrupted by cool rainy weather. A strong wind disturbs the flying of the insects. In hot windy weather, though images were found in considerable numbers in the fruit-trees, only a few of them were caught by the traps. The blue dish efficiently collected various *Hymenoptera* species occurring in the orchard too.

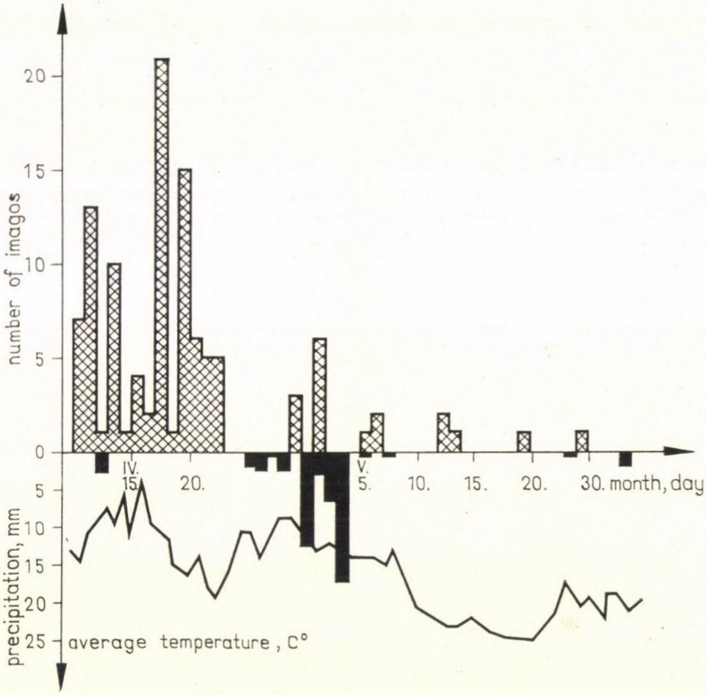

Fig. 3. Swarming trend of *Epicometis hirta* Poda imagos in a homeplot orchard on the basis of six traps, in 1971

Conclusions

The blue colour-trap used for studying the swarming dynamics of *E. hirta* imagos has further increased the number of means of forecast. According to our pilot experiments trapping was not considerably influenced by the shape and size of the traps. This point as well as the effect of the height above ground at which the traps are placed, and of the shades of colour require further studies. Trapping is considerably influenced by the microecological conditions, therefore in a study of *E. hirta* infestation in a given plant stand at least 5—10 dishes should be used optimally. Further investigations are required to find out whether this method can be used in cotrolling *E. hirta* through the decrease of individual density, and in determining the number of insects per unit area.

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EFFECT OF LIGHT INTENSITY ON PRODUCTION OF TOMATO PLANTS (LYCOPERSICUM ESCULENTUM MILL.)

By

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The chlorophyll content, dry weight, carbohydrate and nitrogen accumulation were investigated in two varieties of tomato in the field at two different light intensities. The higher intensity increased the dry weight, the chlorophyll and carbohydrate concentration but decreased the nitrogen concentration in the plants. "Kecskeméti konzerv" and "Kecskeméti törpe" showed a better relative production at the lowest light intensity provided by shading.

Introduction

The upper limit of crop yields corresponds to the conversion of light energy in the plant material of an order of 1 to 5 percent of the incident light (Nichiporovich 1967). Concerning the utilization of solar radiation much attention has been given to its efficiency. Went (1957) obtained an efficiency of ligth utilization of approximately one tenth of that of full sunlight. Loomis—Williams (1963) observed that the photosynthetic efficiencies of crop surfaces diminished with increasing light intensity, but most crops are capable of utilizing more light than the light commonly available. Kumura (1968) observed in soybeans that the photosynthetic efficiency at a higher light intensity was lower than in shaded habitats.

These experiments were carried out to study the influence of light utilized for the growth and organic matter production of tomato plants grown under field conditions in Hungary.

Material and Method

Two tomato varieties were selected "Kecskeméti konzerv" and "Kecskeméti törpe" due to their superior yielding capacity. The seedlings were grown in the dark in a cool house for four weeks up to transplantation.

The production was compared under two kinds of light regime. The high light intensity was natural daylight and the reduction of intensity was achieved by suspending wooden frames in a horizontal plane above the plants, covered by perforated reeds. Light intensity was messured two times, in July and August, with a lux-meter Type 1—0 16 USSR in half hour intervals from 6 a.m. till 6 p.m. The average light intensities were 20,000 lux and 4,000 lux for sunlight and shade respectively. The average temperatures under full sunlight were 29—31 °C and 24—25 °C under shading. The treatments were applied in four replicates randomly designed (Snedecor 1956) and the results were statistically analysed.

Samples were collected at the end of August two months after transplantations when the plants were in full vegetative growth. Five plants were selected at random from each plot and the shoots were analysed. The leaves and stemps were separated and dried at 70 °C until constant weight.

The leaf area was estimated with a photo-electric instrument constructed in this laboratory in vivo. The absorption spectra of the leaves were measured with a UNICAM SP 800 recording spectrophotometer. The determination of the chlorophyll content was carried out in 80 per cent acetone according to (VERNON 1960). The total carbohydrata (DUBOIS *et al.* 1956), total nitrogen (LANG 1958) and protein (LOWRY *et al.* 1951) contents were also determined.

Results

It was observed for both varieties that different intensities of illumination applied in our experiments exerted no significant effect either on plant height or leaf area, in the same manner, as in the experiments of BEAN (1964).

The concentration of chlorophyll was found to be higher in shaded plants for both varieties (Table 1). The amount of chlorophyll per plant was changed by shading in an opposite manner grown the wheat plant due to differences of leaf area and thickness (FRIEND 1961). Similar results were obtained in Chlorella (REGER 1965). The ratio of clorophyll-a, chlorophyll-b for both varieties was higher in plants grown under full sunlight and lower in the shaded ones. These differences were not very pronounced as in the case of Fagus leaves (LICHTENTHALER 1969).

Table 1

Chlorophyll content of tomato leaves grown under different intensities of illuminations

Light intensity (Klux)	Chlorophyll mg/g fresh wt.		(a + b) mg/plant		Chloro-a/chloro-b	
	K	T	K	T	K	T
20	1.32	1.44	69	88	3.17	2.75
4	2.60	2.51	118	56	2.59	2.44
LSD 5%	0.60		30		0.41	

for light intensity
K = variety "Kecskeméti konzerv"
T = variety "Kecskeméti törpe"

The dry weight of leaves for both varieties was significantly higher in plants grown under full sunlight compared to the shaded ones (Table 2). This indicates that under our experimental conditions light was the main limiting factor, other factors being favourable as in the experiments (BONDE

1955) in tomato and cocklebur plants. The effect of shading was more pronounced in the variety Törpe. This observation is not in agreement with the experiments in *Lolium perenne* L. (ALBERDA 1965) indicating that the effect of light intensity is greater on plants with a high production level. The dry matter production of the leaves was less effectively influenced by shading than the dry matter production of the stems. This may be due to interactions between the microclimatic conditions, viz. temperature and light.

Table 2

Dry weight of tomato plants grown under different intensities of illuminations

Light intensity (Klux)	Leaf		Stem		Leaf/stem		Total	
	K	T	K	T	K	T	K	T
20	10.0	9.2	13.7	7.2	0.8	1.3	23.7	16.4
4	6.5	3.2	4.0	2.2	1.6	1.5	10.5	5.4
LSD 5%	2.5		4.1				4.3	

for light intensity
K = variety "Kecskeméti konzerv"
T = variety "Kecskeméti törpe"

Table 3

Carbohydrate and nitrogen content of tomato plants grown under different light intensities

Light intensity (Klux)	Carbohydrate mg/g dry wt.				Nitrogen mg/g dry wt.				Carbohydrate/Nitrogen			
	Leaf		Stem		Leaf		Stem		Leaf		Stem	
	K	T	K	T	K	T	K	T	K	T	K	T
20	207	86	165	109	28	33	16	13	7.4	2.7	10.3	8.4
4	71	73	118	100	42	45	17	18	1.7	1.6	6.9	5.6
LSD 5%	17		18		5		n.s					

for light intensity
K = variety "Kecskeméti konzerv"
T = variety "Kecskeméti törpe"

The total carbohydrate is much higher in plants grown under sunlight then grown in the shade (Table 3) as in *Lolium perenne* L. (ALBERDA 1965), and Acer leaves (NISHIDA 1962). The nitrogen concentration was also effected by different light intensities in the leaves and stems. Significantly lower concentrations were found in non shaded plants than shaded ones for both varieties. NISHIDA (1962), ALBERDA (1965) and JOHNSON *et al.* (1969) reported similar results with various plant species. It is to be noted that in the leaves developed under low light intensities absolute concentrations of nitrogen compounds surpassed the values obtained at higher light intensities. The ratio of carbohydrate and nitrogen containing compounds were higher in under intensive illuminations. The data suggest the presence of some varietial difference in relative carbohydrate accumulation, the ratio of carbohydrate (nitrogen for "Kecskeméti Konzerv" being higher in each experimental plot in each treatment.

Table

Utilization of solar energy under field conditions by tomato plants

Light intensity (Klux)	Konzerv	Törpe
Dry weight per plant/Klux		
20	1.18	0.82
4	2.64	1.38
Leaf area per plant/Klux		
20	0.45	0.76
4	2.92	1.68
Chlorophyll per plant/Klux		
20	3.45	4.40
4	29.50	14.00
Carbohydrate per plant/Klux		
20	0.22	0.08
4	0.23	0.11
Leaf protein per plant/Klux		
20	0.09	0.12
4	0.58	1.22

Table 4 presents indexes for the relative estimation of the utilization of light energy calculated for dry weight to leaf area, chlorophyll, carbohydrate and leaf protein per plant. The efficiencies of utilizing solar energy at different intensities for both varieties of tomato plants were higher in the lowest intensities provided by shade as in *Dactylis glomerata* (BEAN 1964) than at a low level of light intensity at which chloroplasts become light saturated which is the major factor leading to low photosynthetic efficiency at a high light intensity. The in vivo absorption of shaded crops for both varieties were also found to be a little more than the crops grown under sunlight (Fig. 1). NICHIPOROVICH

(1962) in an experiment in maize demonstrated that due to the proper utilization of the optical capacity of the plant in evenly distributed light, the efficiency of light utilization apparently increased with a decrease of light intensity. Kok (1969) also explained that the virtually complete absorption of incident radiation by a leaf or a dense culture of algae results in a strong internal intensity gradient. Directly exposed chlorophlasts may be light saturated, others receiving less light use it more effectively while those, deep in the tissue, may be in virtual darkness. As a result, such a system approaches light saturation much more gradually than an optically thin layer of algae or chloroplasts. Furthermore directly exposed chloroplasts may be light saturated, while others receiving less light use it more effectively.

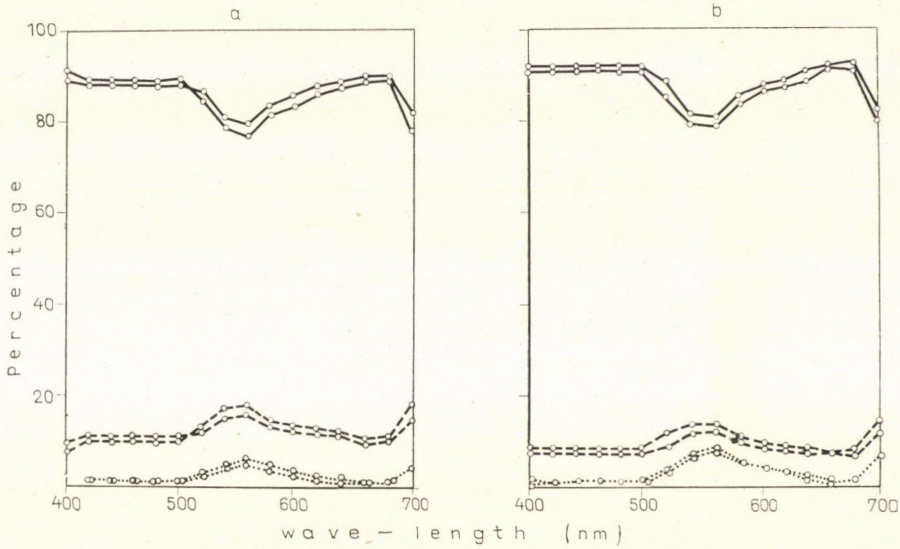

Fig. 1. Showing in vivo absorptio, reflection and transmission of tomato plants grown under different illumitation intensities. a) Under sunlight, b) under shade. ●——● Konzerv, ○——○ Törpe, Absorption; ●------● Konzerv, ○------○ Törpe, Reflection; ●········● Konzerv, ○········○ Törpe Transmission

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VARIA

CUCUMBER VARIETY KECSKEMÉTI HAMVAS

Taxonomical place: *Cucumis sativus* L.

Origin: developed from a local variety in the district of Kecskemét.

Beginning of breeding: 1948.

State qualification: variety licensed for circulation 1951, state certified variety 1969.

Breeder: Mrs. Lajos Kőrös, Kecskemét.

General characterization: early variety with medium long, stumpy, cylindrical white fruits; fairly resistant to diseases. Good dessert cucumber, suitable for preservation.

Morphological description:

Root system: strong, well developed

Shoot system: of vigorous growth, moderately branching

Stem: costate-angular, bristled, light yellowish-green

Foliage: medium dense; leaves are yellowish-green, hairy, palmate with 3—5 lobes

Flowers: with light sulphur-yellow corolla

Fruit: medium long, light green (white when ripe), stumpy cylindrical, with a triangular cross-section and large pulp. On the surface of the fruit white bristles can be seen

Seed: oblong egg-shaped, flat, light yellowish-green; 1000 seed-weight: 18 g.

Biological character:

Vegetation period: from sowing to ripeness 80—85 days, generally short (BALÁZS—FILIUS 1970).

Water requirement: moderate; drought tolerant even in sand soils (Tuza 1971)

Resistance to diseases: not susceptible to fungi (Balázs 1970)

Farm technology requirements:

Sowing: at the beginning of May

Soil requirement: not particular; productive even in light soils

Productivity: 196—422 q/ha of which first class fruit is 41—70 percent, depending on the circumstances; the highest yield is harvested in the third-, fourth-, sometimes fifth week of the ripening period, which means a 20 percent picking per week (Tuza 1965, 1971). The fruits have a high processing value.

Region of cultivation: mainly in the district of Kecskemét (where it is wide-spread); it is successfully grown, however, in other parts of the country too.

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IS THE BRUNNER—ANTONI METHOD SUITABLE FOR THE DETERMINATION OF THE AUXIN CONTENT IN PLANT TISSUES?

The method suggested by Brunner—Antoni (1971) is really rapid and simple, giving repeatable results with a given plant material. However, some doubts arise with regard to its general applicability. Therefore, some questionable points of the method will be discussed below.

Using the recommended method, in 2 grams of somewhat senescent but healthy apple leaves we found as many as 11 μg of indoleacetic acid (IAA) after subtracting the O.D. value of the perchloric acid (PCA) extract from that of the Gordon—Weber reagent (GWR) extract, and reading the corresponding IAA quantity from a calibration curve. Thus, our leaves should have contained $500 \times 11\ \mu g = 5500\ \mu g$ of IAA in kg fresh weight. This amount of IAA seems to be clearly overestimated, taking into consideration that vegetative green plant parts with the exception of shoot apices usually do not contain more than one hundred micrograms of IAA per kg fresh weight (Leopold 1955).

Furthermore, it is known that the colour intensity of the IAA chromophore decreases after 35 min. incubation in aqueous solutions (Gordon—Paleg 1957). Consequently, it is highly probable that the IAA found in apple leaves with the Brunner—Antoni method is not an indolic compound at all.

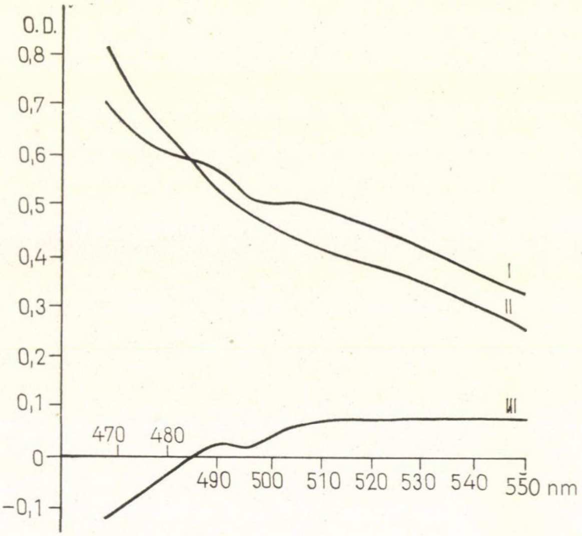

Fig. 1. Light absorption curves of Gordon—Weber reagent extracts (I), perchloric acid extracts (II) of apple leaves made according to Brunner—Antoni (1971), and the difference spectral curve (III) derived from both the former absorption curves

To test this probability, the light absorption spectrum of GWR extracts made from apple leaves according to Brunner and Antoni was investigated in the visible range between 470 and 550 nm. A difference spectrum of GWR and PCA extracts was also constructed. As seen on Fig. 1, there is no maximum at 510 nm (Nitsch 1956) on either the GWR extract or the difference curves. This is not surprising, because our extracts were always medium to deep brown and the characteristic purple-red IAA colour never appeared.

Moreover, when an extract volume representing 0.2 g of leaves (equivalent with 1.1 μg of IAA) was applied on silicagel G (Merck) thin layers, chromatographed with isopropanol-ammonia-water (80 : 5 : 15) as solvent in the dark at room temperature, and developed subsequently with GWR or Procházka reagent (Procházka 1958), we did not obtain any indolic bands on our plates, although the 1.1 μg of IAA exceeds the lower sensitivity limit of both developers on thin layer considerably (Collet—Dubouchet—Pilet 1964). In addition, the material eluted from the layers at the region of IAA with methanol did not give a UV absorption curve resembling that of IAA.

It is possible that the Brunner—Antoni method can produce acceptable results under special circumstances, e.g. when the IAA content of plant tissues free from iron-reacting, PCA-soluble substances is to be measured. However, it is clear from the above observations that the GWR method is not suitable for direct indole auxin determinations in general. This has already been pointed out by Platt—Thimann (1956), as well as Gordon—Paleg (1957), too.

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EFFECT OF METEOROLOGICAL FACTORS ON THE YIELD OF WINTER WHEAT AT MARTONVÁSÁR

The amount of yield is the result of the joint effect of a number of edaphic, climatic and agrotechnical factors. The climatic factors have both indirect and direct effects and considerably influence the course of development in wheat which in turn affects the amount of yield.

LELLEY—RAJHÁTHY (1955), LUKYANENKO (1965) consider the characteristics of the variety, while LEGÁNY (1931), KOVÁTS (1960), KOLTAY (1962) the effect of agrotechnical factors to be decisive.

AZZI (1927), BERÉNYI (1951), PRIKRYL (1965), WILLIAMS—ROBERTSON (1965) and ULANOVA (1965) attach primary importance to precipitation, while PINTÉR (1955), BACSÓ (1963) and MIHÁLYFALVI (1971) to temperature and sunshine hours, or their joint effect.

According to DÉGEN (1931), KREYBIG (1953), BERÉNYI (1958) edaphic, climatic and agrotechnical factors jointly determine the amount of yield.

Under the continental climate of Hungary dry and wet years alternate, and the distribution of precipitation is uneven. It is important to know the effects exerted by the climatic factors on the wheat varieties. The present paper analyses these effects in the period that has passed since the varieties Bezostaya 1 and Fertődi 293 were introduced.

Our investigations were carried out between 1960 and 1970 at Martonvásár, at the Agricultural Research Institute of the Hungarian Academy of Sciences with the wheat varieties Bezostaya 1 and Fertődi 293.

The soil of the trial area was a medium heavy fertile clay with a less than average capacity to retain phosphoric acid and water. Wheat was grown under good agrotechnical conditions in a random block or balanced lattice design with six replications. Plot size varied between 20 and 27 m^2. Sowing took place in the second week of October.

Previous crops were summer barley in the first two years and later peas with sunflower. Every three years 500 q/ha farmyard manure, and every year 120 kg N-, 100 kg P_2O_5- and 80 kg K active agent were distributed on the plots.

In the experimental period most of the years — though highly different from one another — were rainier than the 40 years average (Table 1).

Table 1

Yield and the meteorological data examined
(Martonvásár, 1960—1970)

Year	Yield (q/ha)		Precipitation mm		Temperature °C		Sunshine hours	
	Bezostaya 1	Fertődi 293	crop year	calendar year	crop year	calendar year	crop year	calendar year
1960	50.93	50.68	576	706	9.6	10.5	1738	1911
1961	66.43	62.97	583	501	10.6	11.7	1788	2210
1962	58.12	55.67	451	478	9.5	10.5	1685	1985
1963	31.34	30.01	773	654	8.4	8.7	1900	2173
1964	34.77	31.14	523	674	8.9	10.3	1707	1806
1965	57.12	57.29	875	837	9.0	9.9	1537	1968
1966	45.87	44.14	743	760	9.9	10.8	1760	1974
1967	55.70	50.27	663	507	10.8	11.8	1809	2114
1968	69.56	68.28	472	542	10.7	11.4	1836	1930
1969	51.55	49.37	642	677	9.6	10.5	1559	1769
1970	41.72	39.96	666	605	9.5	10.3	1419	1618

Our investigations concerned the correlation between the productivity of the varieties Bezostaya 1 and Fertődi 293 on the one hand, and precipitation, temperature and the number of sunshine hours on the other. The three climatic factors were broken down to crop year (1st September—31st July) and calendar year (1st January—31st December). Correlation between meteorological elements and yield was studied on the basis of annual and monthly averages.

Relationship between yield and precipitation. Correlation coefficients between the meteorological factors examined and the yield are shown in Table 2. The correlation coefficients of both varieties show a medium close negative correlation between precipitation in both calendar and crop years and yield. On this basis the varieties examined can be grown more successfully under drier conditions.

According to Fig. 1a correlation between monthly total precipitation and yield shows the same trend in Bezostaya 1 as in Fertődi 293. In almost every case the correlation coefficients are of the same order and sign.

Rain in September — which otherwise influences the preparation of seed beds and provides a certain amount of water reserve for germination — had no practical effect on yield. There was a moderately close positive correlation shown between October precipitation and the average yield of wheat. Between precipitation in November, December, January, February, March and yield a fairly close negative correlation was found. This suggests that abundant winter precipitation may have an adverse influence on the amount of yield.

The high positive effect of April and May rains is well known and proved by our investigations too. Moisture in June and July again showed a negative correlation with yield.

Relationship between yield and temperature. Between the annual mean temperature and the yields of the wheat varieties examined a medium close or close positive correlation was found (Table 2). This suggests that between certain limits wheat gives a positive response to temperature.

Correlation between the monthly temperature means and yield varied in the different months, nevertheless the varieties examined gave the same response to this meteorological

Table 2

Correlation coefficients between yield and meteorological data
(Martonvásár, 1960—1970)

Designation	Period	Bezostaya 1	Fertődi 293
Yield-precipitation	calendar year	—0.4032	—0.3179
	crop year	—0.3353	—0.2910
Yield-temperature	calendar year	+0.7251	+0.6497
	crop year	+0.7643	+0.8067
Yield-sunshine hours	calendar year	+0.2608	+0.2300
	crop year	+0.0835	+0.0499

factor too (Fig. 1b). Air temperature in September and November had practically no effect on the yield of wheat. In October, as well as in the winter and early spring months the heat requirement of wheat is rather high, with a peak in January. Temperature in May and June has little role in influencing the yield, while July temperature showed a slight negative correlation.

Relationship between yield and sunshine hours. On the basis of calendar- and crop year data there was but a slight — if any — correlation between the annual total of sunshine hours and yield (Table 2). When, however, correlation between the monthly totals of sunshine hours

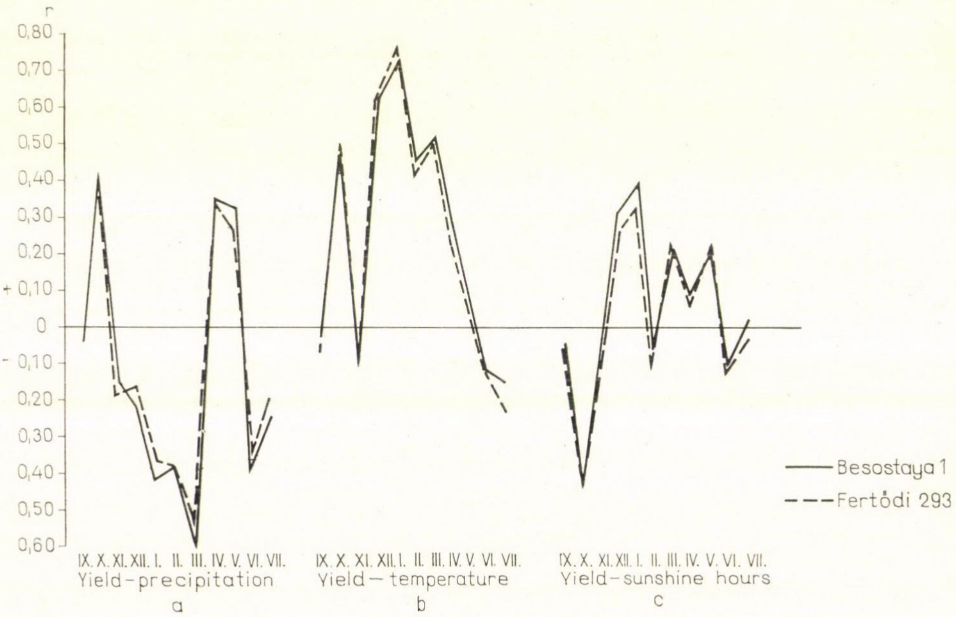

Fig. 1. **Correlation between yield and meteorological data. Martonvásár 1960—1970.**

and yield is considered, interesting data are obtained (Fig. 1). No significant relationship was found between the number of sunshine hours in September, November, February, June, July and the yield. The relationship between the yield and the number of sunshine hours in October was moderately close and negative. It was remarkable that on an eleven years average wheat was found to be indifferent to sunshine in November, at the same time sunshine requirements appeared to increase in December and January. Correlation between the number of sunshine hours in March, April, May and the yield showed a slight positive trend.

As for the relationship between yield and precipitation, temperature and sunshine hours, no difference was found between the two varieties.

From the correlation between precipitation, temperature, sunshine hours and yield conclusions can be drawn on the climatic requirements of wheat varieties on the one hand, and on the influence of meteorological elements on the yield, on the other. However, the meteorological factors act jointly, and their effects are often interrelated as well.

According to the results obtained, weather conditions in September and November have no direct effect on the yield. On the contrary, the trends of precipitation, temperature and sunshine hours in October are much more significant. October weather is favourable when it is rainy and warm.

The joint effect of precipitation, temperature and the number of sunshine hours is interesting in the winter months. It seems favourable if the winter is relatively mild, sunny and not too rich in precipitation. Of the three meteorological factors examined temperature exercised the highest influence on yield (in January: $r = 0.76$). It was considerably affected by the trend of precipitation at the end of winter or early in spring too (in March: $r = 0.58$), while under Hungarian conditions the number of sunshine hours had a less important role (in January: $r = 0.39$).

Since in the period examined winter destruction was negligible in the experiments, the negative effect of low winter temperatures could not be attributed to frost damage. Low winter temperatures may have an unfavourable physiological influence acting probably through the weakening and spring regeneration of plants. Winter precipitation includes snow-cover; the thicker it is, the more unfavourable are the conditions of transpiration, the more are the plants cut off from sunshine and the higher the possibility of damages done by fungi (Fusarium). According to our investigations, under Hungarian conditions the influence of weather conditions in winter on the yields of winter wheat varieties is much greater than has been thought so far.

The role of spring weather is better known. Precipitation and sunshine in April and May, as well as higher temperatures in April have a favourable effect. The latter has no decisive role in May, as at that time the temperatre is usually satisfactory for the normal development of wheat. It is interesting that precipitation, high temperature and sunshine in June influence the yield adversely. Correlations between July data and yield are relatively poor.

In conclusion, the factors that satisfy the demands of plants the least in a given period of development are the most decisive from the point of view of yield.

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THE ROLE OF KEEL IN THE AUTOMATIC DEHISCENCE OF LUCERNE (MEDICAGO SATIVA L.) FLOWER

Researchers dealing with the biology of the flowering of lucerna pointed out that the dehiscence of the flowers that occurs automatically either under external influence of for physiological reasons -- is indispensable for pollination (Heszky 1960). In the flower two opposed forces are in interaction. One of them is the pressure caused by the tendency of the staminal column to curve, the other is the resistance of the keel blades to the staminal column. It can be supposed that the automatic dehiscence of the flower is caused by the cessation of the balance between the forces which is explained by the researchers in different ways (Heszky 1971). The contradiction between the results of the investigations is caused — according to Larkin—Graumann (1954) — by the fact that the simultaneous existence of the two forces prevents the separate and precise assessment of either of them.

By our method of preparation we succeeded in removing the staminal column from the keel without separating the blades of the keel. We cut through with a scalpel the claw of the keel in the flower, from which organs not taking part in closing the flower had been previously removed (HESZKY 1972) (Fig. 1). After the cutting the staminal column curved without the dehiscence of the keel (Fig. 2). Then the staminal column was carefully pulled out of the keel (Fig. 3), so that its two blades did not separate (Fig. 4). In a similar way as described above the staminal column was also preparated from flowers from which only the calyx and the standard had been previously removed (Fig. 5).

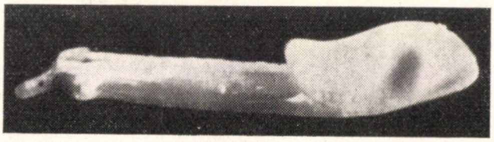

Fig. 1. The closed keel with the staminal column after the removal of calyx, standard, blades of wings, horn-like- and small projections of wings, from side-view

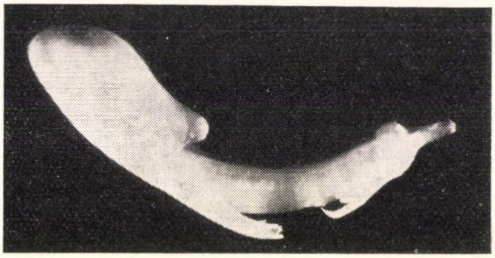

Fig. 2. Curving of staminal column without the dehiscence of the keel after cutting through claw of the kell, from side-view

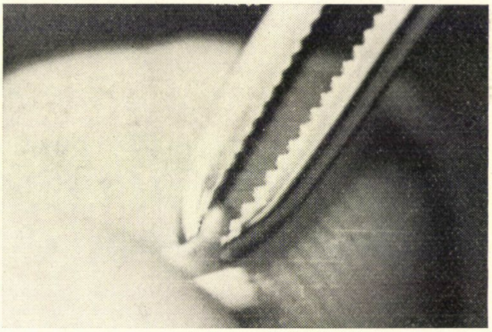

Fig. 3. Removal of staminal column from the closed keel, from side-view

By this method of preparation the separate study of the staminal column on the one hand, and of the closed keel on the other, became possible. If the closed empty keel was placed into warm water, or the connection between the blades loosened with a pin, or else the keel heated with a hand-magnifier, in each case the sudden, explosionlike tripping of the blades of the keel was observed (Fig. 6). An automatic tripping of the closed keels from which the staminal column had been removed occurred after 50—60 sec. in water of 55—58 °C, and immediately in 80—90 °C water — depending on the variety.

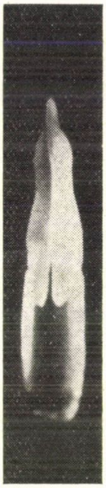

Fig. 4. The closed empty keel after the removal of the staminal column, from above

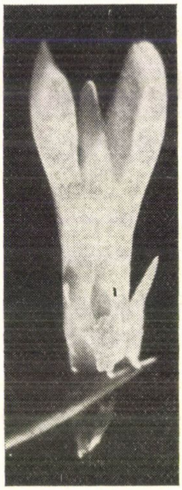

Fig. 5. The closed keel with the blades and projections of wings after the removal of staminal column, from above

Fig. 6. Automatically dehisced empty keel, from above

These results suggest that the immediate cause of automatic dehiscence is the tripping of the keel. Changes in the osmotic pressure of the staminal column, climatic conditions, the shape of the horn-like projections of the keel blades, etc. — are all only indirect causes of automatic dehiscence. Changes in these indirect causes always lead to the same result: the connection between the two closed blades of the keel becomes loose. In the case of automatic dehiscence — under the influence of the indirect causes — the connection between the blades of the closed keel becomes so loose that the keel bursts open by itself.

It can be supposed that the extent of automatic dehiscence of closed keels deprived of staminal column will be in the near future a selective character in breeding lucerne varieties with easily dehiscing flowers.

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Prepared at the Institute of Agrobotany, Tápiószele.

L. Heszky

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TESTING OF VARIOUSLY COATED SPRING WHEAT

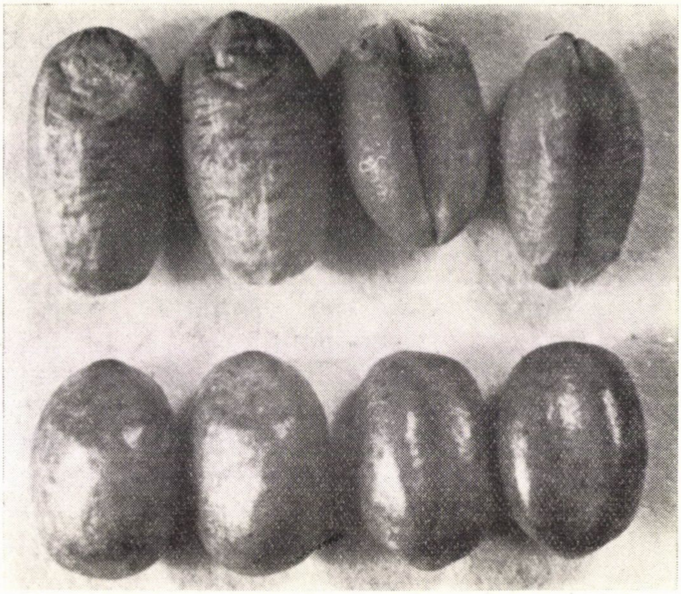

Fig. 1. Above: wheat grains without coating; below: the same with coating

SCHREIBER—LACROIX (1967), researchers of the Department of Plant Science at the University of Manitoba, Winnipeg, have elaborated a method which makes possible the autumn sowing of non-winter-hardy spring wheats or other plants. In Canada where the vegetative period of wheat is short, and in the north the winter is so severe that winter wheat can hardly be grown, the new method is of great importance. Spring wheat sown in autumn emerges earlier than when sown in spring, utilizes winter precipitation better and even its vegetative period will be shorter. The point is to cover the seed with a thin plastic layer which does not let water through, thus the grains do not germinate in autumn. In three months the coating dissolves, and by the time the weather begins to warm up, the wheat can germinate. The method has the great advantage that soil cultivation prior to spring sowing does not dry out the seedbed.

The two researchers later gave the following written information:

In the course of further experiments in 1967 and 1968 they found that

a) the root system of spring wheat varieties sown in autumn is stronger and utilizes water better,

b) spring wheat varieties sown in autumn show better tillering and develop larger spikes,

c) their stalks are shorter and thicker,

d) they yielded 14 percent more in 1967 and 50 percent more in 1968,

e) the total protein content of the grain increased by 1 percent.

In Hungary no spring wheat is grown because winter wheat always yields better. Spring wheat production is not justified even by the destruction of stands sown in autumn, since the winter-hardiness of varieties grown in Hungary is satisfactory. Thus, the experiment was not started for such practical purposes, but to find out whether the proposed method can occasionally be applied under Hungarian conditions in the case of other spring crops.

In the autumn of 1970 Schreiber sent five kinds of spring wheat seed grains. Four samples were covered by coatings of various colour and gloss; one sample was untreated. The composition of the coating was not given. All the five samples originated from the same spring wheat variety. The investigation was aimed at determining to what extent the four types of coating meet the requirements under Hungarian conditions, and what is the difference between treatments sown in autumn and those sown in spring.

The experiment was set up with four replications, on a medium heavy clay soil, with 5 m long single-row plots. The distance between the plots was 50 cm, while between the seed grains 2 cm. Sowing took place on October 8th in autumn, and on February 23rd in spring. The precipitation during the vegetative period is shown in Table 1.

Table 1

Distribution of precipitation during the vegetative period

Monthly amount of precipitation, mm		
1970	October	23.4
	November	44.4
	December	38.0
1971	January	51.2
	February	7.8
	March	35.4
	April	10.4
	May	67.3
	June	40.9
	July	49.1

The distribution of the precipitation was favourable even for the spring sowing. 25—30 days after the autumn sowing all four treatments began to emerge, though scarcely. Since the number of grains sown was known, it could be established that on an average of the four series 21.3, 14.0, 16.1 and 17.1 percent of the grains germinated. In December — owing to the weather — no further counting was possible.

After spring had come emergence continued, however, all the four treatments remained apparently scarce. Owing to the tillering of the plants emerged in autumn and winter, the plants could no longer be counted. Plants emerging in autumn were apparently worn down by the winter, in spite of the fact that the winter of 1970—71 was mild — no temperature below —12 °C was ever measured.

In spring, owing to the highly favourable weather in February, sowing could be carried out very early. Seeds germinated rapidly and the stand developed evenly. Heading began on May 22nd in the case of autumn sowing, and on May 31st with spring sowing. Plants sown in autumn became mature on July 5th, while those sown in spring on July 17th.

The average yields of the plots in the different treatments are shown in Table 2.

According to the table all the four autumn sowing treatments yielded less than the spring sowing, with an error probability of S.D. 0.1%. Further, the first treatment gave a lower yield than treatments 2, 3 and 4, with a probability of S.D. 0.1%. On the other hand, between treatments 2, 3 and 4 no demonstrable difference was found even at a level of S.D. 5%.

Table 2

Average yields of plots in the different treatments

Treatment	Average yield of plot g	Significance	
		related to the standard	between the treatments
1. autumn sowing	214.5	+++	+++
2. autumn sowing	354.0	+++	---
3. autumn sowing	341.2	+++	---
4. autumn sowing	366.3	+++	---
5. spring sowing	549.2		
	S.D. 5%	64.31 g	
	S.D. 0.1%	127.44 g	

Stalk shortening reported as having occurred in Canada in 1967 and 1968 was not observed in the experiment. Even the slight difference in lodging could only be observed, inasmuch as in the treatments the stand was thinner.

On the basis of the experiment it was found that under the weather conditions of the year 1970/71 none of the four coatings provided sufficient protection against germination in autumn. The winter wore down the plants, a part of the seed was destroyed, therefore the stands sown in autumn became thin by spring.

If the value of the protecting coating is judged by the crop result, then the coating of the first treatment is considered to have been the poorest. Coating was not satisfactory in treatments 2, 3 and 4 either, however, between these treatments no significant difference was found.

No considerable stalk shortening or improved lodging resistance could be put to the credit of autumn sowing either. On the other hand, the 12 days shortening of the vegetative period is a highly positive feature.

Thus, sowing the coated seed grain of spring wheat in autumn did not work under Hungarian conditions. The protective coating which under the climatic and soil conditions of Canada proved good, in Hungary did not provide sufficient protection against germination.

It must be noted, that in 1970—71 the possibility of sowing very early in spring and the distribution of spring precipitation were much more favourable than usually. This increased the difference between treated and untreated plots.

No doubt, the idea of coating the seed of certain spring cultures and sowing it in autumn is good, and will be very useful in many places. Coating should be prepared, however, in accordance with the circumstances, which cannot be a difficult task today.

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J. LELLEY

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NEW METHOD FOR AN EARLY AND QUICK DETECTION OF STOCK-SCION INCOMPATIBILITY

(Compatibility ratio)

Transversal sections are made on one-year old grafts 15 mm above the upper border of the place where the stub has been cut off. Then measurements are made on the diameter of the above mentioned transversal section of the scion, in the plane passing through the median of the place of the cut-off stub (Fig. 1).

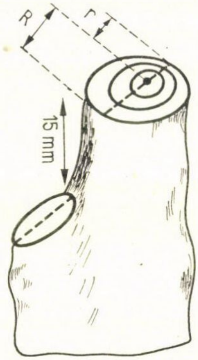

Fig. 1. Pattern of stock-scion compatibility study Sign used: $\frac{R}{r}$ = compatibility ratio. Compatibility: $\frac{R}{r} = 1$; incompatibility: $\frac{R}{r} > 1$

The first diameter measurement is made between the border of the section on the side of the cut-off stub and the central pith. This gives the value of "R". The value of "r" is obtained by measuring the opposite side of the diameter.

The nearer the value of $\frac{R}{r}$ — i.e. of the compatibility ratio — is to 1, the smoother is the upward and downward transportation in the region of the grafting place. On the other hand, in the case of an incompatible stock-scion combination sectorial transport disturbances are indicated by the excentric thickening of the scion above the place of grafting, and — consequently — by an increase in the value of "R" with the simultaneous decrease of the value of "r", which raises the value of R above 1 (e.g. 1.5 etc.). The higher the value of this quotient, the greater the incompatibility indicated.

In similar studies (Brunner 1968, Brunner—Antoni—Gál 1970) with compatible and incompatible stock-scion combinations of apricot-, peach- and pear grafts excentric thickening above the place ofgrafting in incompatible combinations was observed as early as at the age of one year.

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Prepared at the Horticultural Research Institute, Budapest.

T. Brunner

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THE BIRTH OF MYCOLOGY

International scientific co-operation in the 16th century

Shakespeare said that ignorance was plague and science was protection.[1] Well, this is especially true as far as mushrooms are concerned, as mushroom poisoning demanded many victims, and the tragedies were caused in almost every case — except wilful murders — by ignorance.

Still, what a long time had to elapse until mycology, the science dealing with mushrooms was born. The historians of science pointed out beyond any shadow of doubt that mycology as a science was born in Hungary. There are authors who say that all this is due to the Hungarian hospitality. If this were true, it is worth paying attention to the fact: what a blessing simple genuine hospitality may bring on the whole human race. However, as you will see, at the birthplace of mycology not only Hungarian hospitality but Hungary's devotion to science too was present.

Description of the age and Clusius' presentation

The age we have to go back to is the 16th century when Hungary was divided into three. The centre of the country was under Turkish rule, the western part lived in continuous fighting, the east in compromise. The centre of Hungary which, with its beauty and richness had once amazed even the envoys of the great European powers, was ruined, and the greatest part of the country was inaccessible to scientific research. It remained "terra incognita" for several generations.

The 16th century had three great botanists: Dodoneus (1517—1585), Clusius (1526—1609) and Lobelius (1538—1616). It was Clusius who visited Hungary; he was in the western and northern borderland. And even though he did not stay for long, on the basis of his work and with the collaboration of his Hungarian friends, mycology and the Hungarian botany were born.

Jules Charles de l'Escluset, or as he called himself according to the fashion of the age: Carolus Clusius (1526—1609), the first to describe the Spanish and Hungarian flora, the founder of mycology need not be presented. Linné mentions him among the best specialists in descriptive botany. Sprengel calls him the martyr of science, the greatest and worthiest plant researcher. According to Cuvier he was the most educated man of his century. Planchon calls him aprince of the 16th century descriptive natural science. According to Neilreich he was a phenomenon who appeared as a sparkling meteor.[2]

The scientists of Hungary used the most diversified epithets when discussing the activity of Clusius. According to Sadler, Clusius was the famous founder of the scientific stage of Hun-

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[1] "...ignorance is the curse of God, Knowledge the wing wherewith we fly to heaven." Second part of King Henry VI; IV. 7.

[2] Istvánffi: Clusius mint a magyar gombászat megalapítója (Clusius as the founder of Hungarian mycology). Budapest, 1895. p. 272.

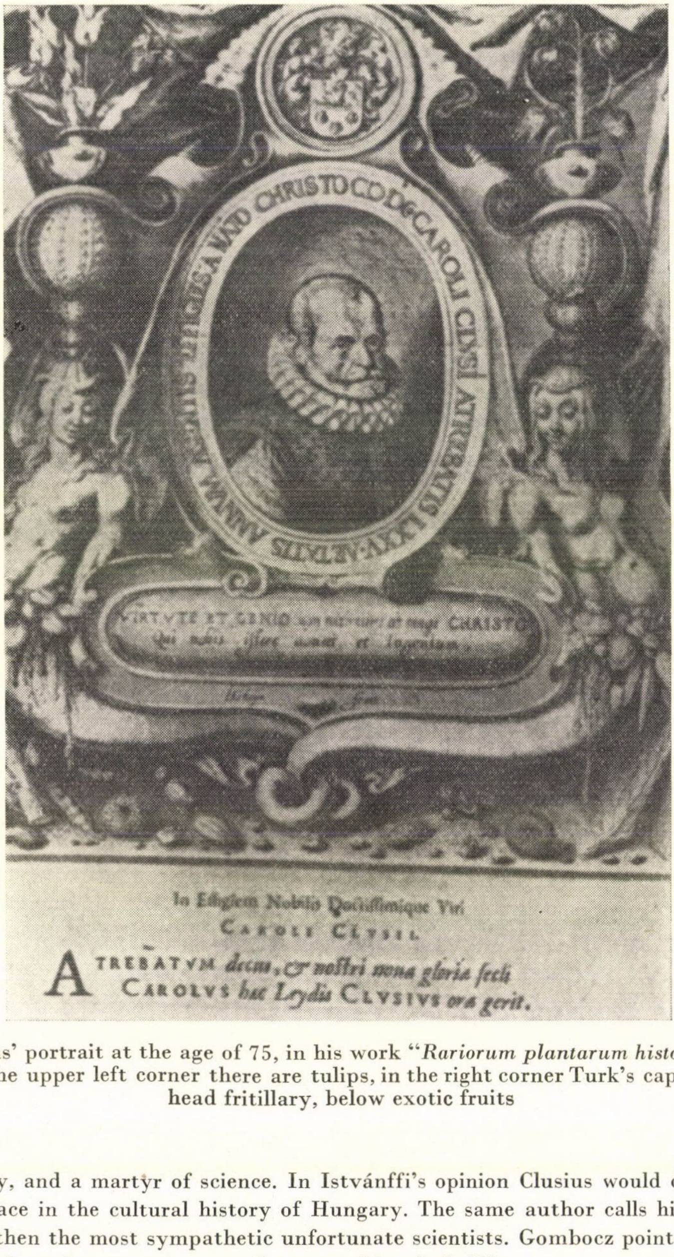

Fig. 1. Clusius' portrait at the age of 75, in his work "*Rariorum plantarum historia*" published in 1601. In the upper left corner there are tulips, in the right corner Turk's cap, and a snake's head fritillary, below exotic fruits

garian botany, and a martyr of science. In Istvánffi's opinion Clusius would deserve a more important place in the cultural history of Hungary. The same author calls him Linnaeus of mushrooms, then the most sympathetic unfortunate scientists. Gombocz points out that Clusius' botanical work was as extraordinary as his whole life.

It is not the first occasion the Hungarian history of science pays the tribute of admiration to Clusius' memory. Some maintain that perhaps nothing new can be said about him. Among the numerous articles and papers the most important ones are referred to here as sources; even the final identification of mushrooms described by Clusius was the merit of a Hungarian scientist, Gyula Istvánffi.

Clusius' Hungarian friends

They are not inaptly called by Gombocz the members of the "small Németújvár* circle of scientists". Boldizsár Batthyány (1538—1590), a Hungarian aristocrat of classical education, one of the most interesting figures of Hungarian science- and cultural history, was the host. He played an important role in fighting against the Turks. For his military merits he was appointed Warden of the King. And the fact that he spoke six languages besides his mother language was made known by Clusius himself. ("Namely, this brave man speaks Latin, Italian, French, Spanish, German and Croatian perfectly, besides Hungarian.") Batthyány remained Clusius' faithful ally to the last. It was in his court that Clusius made friends with István Beythe (1532—1612), court-chaplain of the Calvinist Batthyány who accompanied Clusius on his collecting tours, and helped him in other ways too. The *Stirpium Nomenclator Pannonicus* is their joint work.

In his writings Clusius also referred to Sambucus, János Zsámboki (1531—1584), the historian, physician, cartographer, and book- and art collector of European fame. In Pozsony he was always the guest of György Purkircher (1530—1578), physician, after whom he named the American bean variety *Phaseolus Purkircheri*, as he gave it to him. With Miklós Isthvánffy (1538—1615), the famous politician and historian he maintained a close friendship and frequent correspondence up to his death. The library of the Leyden university keeps eight letters written by Isthvánffy to Clusius.[3]

Of course, the above list does not contain every place where Clusius stayed in Hungary, since he visited many places during his collecting tours. In his works he mentions Dévény, Stomfa, Pozsony, Szent-György, Nagyszombat, Magyaróvár, Sopron, Pinkafő, Velem, Rohonc, Németújvár, Körmend, Ó-Szalónak, Körtvélyes, Alsó-Lendva, Csesztreg, Csáktornya, Grebén, Peklenicza, Strukovecz and Varasd by name.

The result of the above friendships and collecting tours was a paper entitled *Fungorum in Pannonia observatorum brevis historia* published in Antwerpen, 1601, on pages 261—268 of his work *Rariorum plantarum historia* . . . This is the oldest known mycological work. It is this the so called Leyden Clusius-Codex is connected with.

Fungorum historia

Before Clusius mushrooms were dealt with only incidentally. Bock discusses 12 mushrooms; Matthiolus mentions some species of his country in addition to those described by Dioscorides, but the other botanists of the 16th century, together with them, hardly speak of 40—50 mushrooms. It is still more important that until Clusius nobody ever tried to systematize the mushrooms of some area searched through.

In the *Fungorum historia* Clusius thoughtfully describes the edible and poisonous mushrooms of the western part of Hungary. He gives their occurrence and Hungarian names and presents their lifelike pictures.

Clusius accepted Plinius' classification into edible and poisonous mushrooms. He too set up two large groups and described 21 genus of edible and 26 of poisonous mushrooms, with 46 and 59 species respectively, thus placing a total of 105 species in the two genus. These mushrooms are listed in Gombocz' book "The history of Hungarian botany" (A magyar botanika története) published in 1936 by the Hungarian Academy of Sciences.

Clusius' mushrooms could not be identified for a very long time, although the inter-

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* Small town in the western borderland of Hungary.

[3] These were published by Istvánffi: A Clusius-Codex Mykológiai méltatása (Mycological appreciation of the Clusius-Codex), p. 210—214.

Fig. 2. Boldizsár de Batthyány (d'après son portrait au chateau de Körmend). (After his portrait in the castle of Körmend)

preters were — as Istvánffi put it — a "legion". Fries, Kicke, Brizelmayer, Sadler, all are great names in this special branch of science. Sadler says that only those who are sufficiently expert in mycology can "throw some light" upon Clusius' work. It was he who mentioned István Máttyus who, in his work "Old and new dietetics" (Ó és új Dietetika) between 1787 and 1793 tried to identify Clusius' mushrooms with plenty of success. He also called attention to the following works as being considered commentaries on Clusius' Hungarian mushrooms: I. Sigism, Valent. Popowitsch, Untersuchung vom Meere, Nürnberg 1750. (Sadler added: "Who would have ever thought of finding an explanation of Clusius' mushrooms, and the distinction first pointed out between *Amanita* and *Agaricus* in this book.") — *De fungis officinalibus*, Diss. Leidae, 1702. The author is I. Phil. Breynio; and finally A. G. Bauhin, Pinax. Sectio V. *Fungi* *et tubera.*[4] After Sadler Kalchbrenner set to work, but only Istvánffi succeeded in identifying the mushrooms described by Clusius with the aid of the Clusius-Codex. He identifies the mushrooms listed by Clusius with 86 species of which relatively few are doubtful. This number of species is so high that Western-Hungary can be considered the macromycologically most thoroughly searched area of those times.

Clusius-Codex

While collecting his material Clusius thought of the characteristic colours of mushrooms that no description could substitute for; therefore they had to be depicted. Batthyány was ready to finance the plan, so Clusius went to all lengths until finally he found a suitable painter in Paris. However, the painter worked for the King and could come with Clusius to Németújvár only in August 1579. He painted the pictures of the collected samples in the castle of Németújvár. On 13th November 1584 Batthyány informed Clusius with great pleasure that the pictures of mushrooms painted under his supervision were ready, and asked him to visit him for this and other important reasons, as soon as possible.[5]

In this way the aquarelle collection known as Clusius-Codex came into being. It is a book consisting of 87 pages, which illustrates 221 mushrooms painted in water-colour. Clusius himself made the pictures of *Fungorum historia* after these pictures.

Later even Clusius thought this unequalled collection to be lost. So his mushrooms were always discussed on the basis of *Fungorum historia* published in 1601. And here his interpreters encountered great difficulties. Kalchbrenner pointed out that Clusius' descriptions and pictures of mushrooms were imperfect and sometimes even of inferior quality compared to those of other plants. He gave the following explanation for this: Clusius worked in an unknown field as a pioneer. The essential characters of mushroom genus were not yet clarified and the special language was missing too, without which no precise description could be given.[6]

But the Clusius-Codex — to our great pleasure — was not lost. It somehow got into Sterbeeck's possession who copied it, made copperplates of the copy and published them in his mycological work entitled *Theatrum fungorum oft het tooneel der Campernoelinen*, Antwerpen 1675. At one or two places in his book he tells about copying these mushrooms from Clusius' picture book. ("Van dese heeft ons Clusius in sijnen gheschilderden boeck ses verscheyde figuren ghesteldt, van de welcke hier dry met de letter E vertoont worden." 119.) On other occasions, on the other hand, he writes as if they were his own pictures. And since the later interpreters of mushrooms did not compare these pictures with the Clusius-Codex, and did not read Sterbeeck's text with sufficient attention, they believed — even Britzelmayer — that they were interpreting Sterbeeck's original pictures and descriptions of mushrooms.

Meanwhile É. Morren in his work "Charles de l'Escluse, sa vie et ses oeuvres", Liège 1875, wrote about the Clusius-Codex being kept in the Leyden library. It was from here that Gyula Istvánffi borrowed it, copied it with an amazing zeal and accuracy and published it in 1900 in Hungarian—French with an even greater generosity.

As compared to the other interpreters, he took quite a different view. He took the pictures of the Codex for a basis, as in his opinion it was the only true source, since it originated directly from living nature examined. From the Codex he compiled Clusius' mushroom flora nd compared it with the Historia. He noted that it had been no easy work. Then he began to

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[4] A növénytan történetei (Histories of botany...), p. 105—106.

[5] From what has been said it is evident that the painter was famous. His name is not yet known, though his work is of standing value. Istvánffi cannot praise enough the pictures even after 300 years.

[6] A magyar gombászat fejlődéséről... (The development of Hungarian mycology...), p. 6.

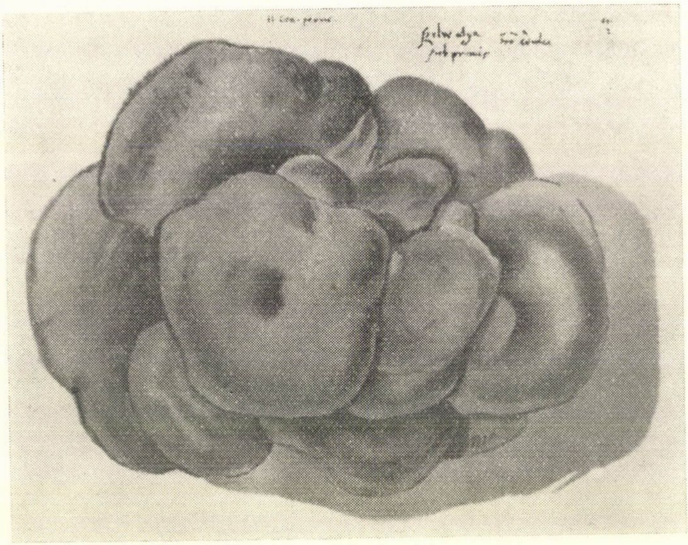

Fig. 3. Table 2. of the Clusius-Coxed. *Pleurotus sapidus* Schulzer. Gen. II. pernic. Szilva alja (Hungarian name)

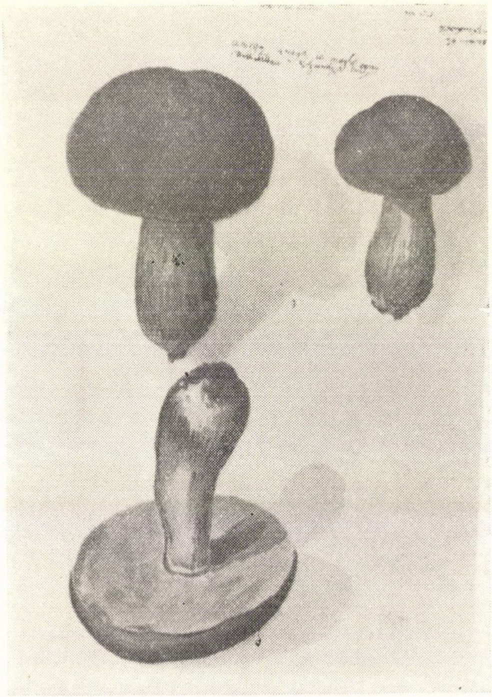

Fig. 4. Table 65. of the Clusius-Codex. *Boletus aereus* Bulliard, Gen. XX. pernic. spec. 3. (varga annya; vargánya) (etymology of the Hungarian name of the mushroom)

insert Sterbeeck's data. Here he did not follow the earlier methods either, as he took the conformity of Sterbeeck's pictures with those of the Codex rather than Sterbeeck's text for a basis. He ranged Sterbeeck's pictures with the corresponding pictures of the Codex; it was only then that the literary data were checked and arranged. So, with thoughtful and meticulous work and great competence he succeeded in the final identification of Clusius' mushrooms. To characterize the work and method of this great Hungarian scientist a short example should stand here.

"*53. — Gen. II. pernic. Cod. 2.*

It grows in spring under the plum trees. I found a variety. Hist. CCIXXVII (p. n. 17).

Under the heading *Gen. pernic. szilw alya sub prunis no edules* the Table 2. II. of the Codex presents (with Clusius' handwriting though, with different pens or at different times) a bushy family consisting of 13—14 mushrooms.

Sterbeeck (p. 243—244. No. 115 A. tab. 27) copied Table 2. of the Cod.

Want alsoo ick de figuren van Clusius in haere volle grootte, met al de coleuren naer het even by my hebbe, soo sal ick de selve volghens haet naer beschreven wesen de Pruym boom navel Fungi veel te samen noemen, — Clusius' pictures illustrate the mushroom in full size and natural colour, and — having it in my possession I shall describe it.

Kalchbrenner 20. II. Szilwa alya = *Polyporus hirsutus* et affines.

Britzelmayr 56. *Polyporus umbellatus* Fr.; St. 27 A. Die Abbildung zeigt die Oberseite eines nicht völlig entfalteten Exemplares. In der Beschreibung sprachen die Angaben über die Hutgrösse, Hutform, Färbung, den Standort und das Substrat dafür, dass die bezeichnete Art gemeint ist. (Kx.: Indétérminé.) Kickx p. 419! Now by Table 2 of the Cod. it can be established that

16. — Gen. VI. escul. Cod. 5.
17. — Gen. VI. escul. Cod. 9.
18. — Gen. VI. escul. Cod. 10. and
53. — Gen. II. pernic. Cod. 2.

are *Pleurotus sapidus* Schulzer, that is a true Hungarian species."

(The photocopy of Table 2. of the Codex is presented in half size.)

We have been at the birthplace of mycology. Even if only in outlines, we have got acquainted with the birth and first results of a new science. And what a science! Namely, while Clusius listed 117 mushrooms, in 1895 some 30,000 species were known already, and today their number may be 100,000. Of course, this number includes the imperfect fungi seen only by microscope. However, these numbers and the great progress must not mislead us. We must not belittle the work Clusius and his Hungarian friends, first of all Boldizsár Batthyány and István Beythe performed. The pioneers' work must not be judged from the aspects of the present times. As a last sentence I cite here the words of Clusius' most competent Hungarian interpreter: ". . . I highly esteem Clusius' extraordinary courage when he dared to summarize, systematize and classify, and on the basis of his own observations, collecting and research work write a History of mushrooms 300 years ago (today it is already 370 years), when the world knew hardly anything of them, and at once conquered the Empire of Mycology . . .".[7]

P. HARGITA

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[7] Istvánffi: A Clusius-Codex... (The Clusius-Codex...). p. 40.

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CHARACTERIZATION OF DIFFERENT DEGREES OF MAGNESIUM SENSITIVITY IN PLANTS

Magnesium accumulates first of all in those plant parts where processes of vital importance take place, such as the seed and the foliage, while less can be found in the stem and roots. Some plants are especially sensitive to magnesium deficiency and are known as deficiency indicators, e.g. the spinach. All this might give the impression that the magnesium content is the highest in the mentioned plant parts and plants. However, often just the opposite can be observed (Table 1, first column). This apparent contradiction gave the idea of determin-

Table 1

Magnesium content and magnesium-coefficient of air-dry plants

No.	Plant	Mg mg%	Mg-coefficient
1	Spinach	100	0.30
2	Soybean shoot	400	0.12
	seed	250	0.17
3	Bean shoot	400	0.15
	seed	160	0.25

ing the magnesium content of plants in such a way as to explain the above. It is for this purpose that the new characterization of magnesium content, the ratio of magnesium content expressed in ion-equivalent to total cation has been elaborated. This quotient is the "magnesium coefficient" (*z*) of which the mathematical formulation is the following:

$$Z = \frac{\mathrm{Mg}}{\mathrm{K} + 1/2\ \mathrm{Ca} + 1/2\ \mathrm{Mg} + \mathrm{Na}}\,.$$

Magnesium content as expressed by the magnesium coefficient is given in the third column of Table 1 which already reflects the earlier statement. The values of the coefficient have been computed for some fifty plants and found to range between 0.1 and 0.3.

On the basis of HOLST's (1967) study on nitrogen uptake we tried to find a correlation between the magnesium coefficient (*z*) and the ion equilibrium constant (*i*). The ion equilibrium constant was derived from the ion-equivalents of four anion (N, P, S, Cl) and four kation (K, Ca, Mg, Na), then macro-elements contained in the plant examined, in the following way:

$$i = \frac{\Sigma^- - \Sigma^+}{2(\Sigma^- + \Sigma^+)}$$

where $\Sigma^- = \mathrm{NO_3} + \mathrm{H_2PO_4} + 1/2\ \mathrm{SO_4} + \mathrm{Cl}$
$\Sigma^+ = \mathrm{K} + 1/2\ \mathrm{Ca} + 1/2\ \mathrm{Mg} + \mathrm{Na}.$

On the basis of analyses made by SVANBERG (1962) the macro-element contents and ion-equilibrium constants of several plants are shown in Table 2.

When plotting the magnesium coefficient against the ion-equilibrium constant the values are found to be situated along three curves (Fig. 1). This grouping is thought to be explainable by the relationship between the sulphur- and magnesium contents of plants expressed by the quotient of the two elements. On the basis of the values of this quotient the plants are classified into three groups: 1. high sulphur content, so called sulphophil plants, 2. normal sulphur content plants and 3. low sulphur content, so called magnesiophil plants. The closeness of the relationship between *z* and *i* as a function of the sulphur content can be proved by an equation too. Equations representing the curves and the respective values of S/Mg are to be found in Table 3.

Table 2

Macroelement content and ion equilibrium constant (i) of several plants

Plant	N	P	S	Cl	K	Ca	Mg	Na	Σ^-	Σ^+	i	z
1. Wheat	5.57	0.47	0.62	0.28	1.27	0.58	0.80	0.10	6.94	2.75	0.22	0.29
2. Rye	5.04	0.46	0.67	0.30	1.38	0.64	0.85	0.10	6.47	2.97	0.18	0.28
3. Barley	4.68	0.45	0.59	0.28	1.31	0.60	0.72	0.10	6.00	2.73	0.19	0.26
4. Oats	5.10	0.41	0.50	0.25	1.29	0.74	0.68	0.14	6.26	2.85	0.18	0.24
5. Timothy	3.43	0.28	0.30	0.45	1.54	0.80	0.43	0.09	4.46	2.85	0.11	0.15
6. Red clover	7.86	0.36	0.44	0.84	2.18	3.50	1.42	0.20	9.50	7.30	0.07	0.19
7. Alfalfa	13.14	0.57	0.70	1.35	3.69	7.20	1.53	0.28	15.75	12.70	0.05	0.12
8. Potato	11.07	0.46	0.91	0.52	5.18	2.65	1.04	0.32	12.96	9.19	0.08	0.11
9. Turnip	13.93	0.87	1.87	1.86	5.38	4.05	1.50	1.70	18.53	12.63	0.10	0.11
10. Sugar beet	11.43	0.74	1.00	2.32	4.10	3.12	2.37	4.20	15.50	13.80	0.03	0.17

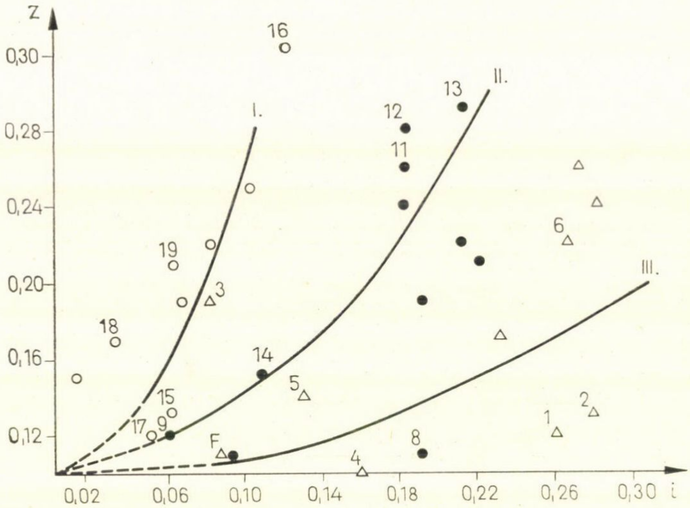

Fig. 1. Relation of the magnesium-coefficient to the ion-equilibrium constant and sulphur-magnesium quotient (I. o magnesiophil S/Mg < 0.7; II. x normal S/Mg = 0.7—1.0; III. Δ sulphophil S/Mg > 1; *i* = ion-equilibrium constant; *z* = magnesium-coefficient)

Table 3

Indices of plant grouping on the basis of sulphur-magnesium quotients

S/Mg	Plant type	Equation of curve	Number of curve
< 0.7	magnesiophil	$17i^2 + 0.1$	I
0.7—1.0	normal	$4i^2 + 0.1$	II
> 1	sulphophil	$i^2 + 0.1$	III

The magnesium-coefficients, ion-equilibrium constants and sulphur-magnesium ratios of several plants are summed up in Table 4 on the basis of analysis data by Holst (1966), Hester—Shelton (1949) and Becker—Dillingen (1933). It was partly on the basis of data in this table that Fig. 1 was made which shows that magnesium sensitivity depends not only on the absolute magnesium content, but on the ratio of magnesium and total cation contents, i.e. the magnesium coefficient, as well as on the sulphur-magnesium quotient too. This is proved also by the results of experiments carried out with a potted sand culture at identical magnesium levels (Table 5). For our investigations we chose three plants with different sulphur- and magnesium requirements.

On the basis of the magnesium-coefficient we would expect that under the same conditions it is wheat that magnesium fertilization has a higher influence on. However, according to our measurements the reaction of radish was better. Namely, the magnesium-coefficient of radish is much above the sulphophil curve, while that of wheat is on the normal curve characteristic of it. It is supposed that the farther the magnesium-coefficient of a plant is from our own curve and the X-axis, the more the plant is sensitive to magnesium deficiency. The enormous increase in the yield of spinach can be explained in two ways. 1. The sulphur-magnesium

Table 4

Magnesium-coefficient (z), ion-equilibrium constant (i) and sulphur-magnesium quotient of various plants

Plant	Σ^-	Mg	z	i	S	S/Mg
1. Cabbage	1.54	0.18	0.12	0.26	1.01	5.6
2. Broccoli	3.84	0.50	0.13	0.28	1.95	3.9
3. Celery	3.52	0.67	0.19	0.08	1.75	2.6
4. Kohlrabi	3.68	0.46	0.10	0.16	1.11	2.4
5. Kale	2.09	0.30	0.14	0.13	0.65	2.2
6. Radish	1.40	0.30	0.23	0.28	0.50	1.5
7. Turnip	12.63	1.50	0.11	0.09	1.87	1.2
8. Lettuce	1.48	0.16	0.11	0.19	0.14	0.87
9. Parsley	1.62	0.19	0.12	0.06	0.17	0.89
10. Potato	9.19	1.04	0.11	0.08	0.91	0.87
11. Barley	2.73	0.72	0.26	0.18	0.59	0.82
12. Rye	2.97	0.85	0.28	0.18	0.67	0.78
13. Wheat	2.75	0.80	0.29	0.21	0.62	0.77
14. Timothy	2.85	0.43	0.15	0.11	0.30	0.70
15. Tomato	15.02	1.92	0.13	0.06	1.25	0.65
16. Spinach	2.6	0.80	0.30	0.12	0.50	0.62
17. Alfalfa	12.70	1.53	0.12	0.05	0.70	0.45
18. Sugar beat	13.80	2.37	0.17	0.03	1.00	0.40
19. Red clover	7.30	1.42	0.19	0.06	0.70	0.31

Table 5

Effect of magnesium fertilization on potted plants

Plant	Z	S/Mg	Yield	CO_2 fixation
			percentage increase by Mg treatments	
Radish	0.22	1.50	17	—
Wheat	0.29	0.77	12	9
Spinach	0.30	0.62	805	11

quotient of spinach is on the border of normal and magnesiophil plants. 2. Inasmuch as we place it in the magnesiophil group, its magnesium sensitivity is understood on the basis of its extremely high magnesium-coefficient. If we place it in the normal group, then the great distance from the curve explains its magnesium sensitivity even better.

On the basis of the above, the introduction of the magnesium-coefficient and sulphur-magnesium quotient to express the magnesium sensitivity seems to be justified and right.

The different degrees of sensitivity to magnesium deficiency in plants were characterized by quotients of equivalent magnesium content and total cation, called magnesium-coefficients. There is a relationship between the magnesium-coefficient (z) and the ion-equilibrium constant (i) which, on the basis of the sulphur-magnesium quotient of the plant, can be represented by three curves. Their equations are:

$$S/Mg > 1; \qquad z = i^2 + 0.1$$
$$S/Mg = 0.7-1.0; \qquad z = 4i^2 + 0.1$$
$$S/Mg < 0.7; \qquad z = 17i^2 + 0.1$$

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AN UNDESCRIBED XEROMORPHIC STRUCTURE IN ARTEMISIA SCOPARIA WALDST. AND KIT.

Artemisia has acquired the representative structural fabrication of a xerophyte to withstand the stresses of drought and desiccation (Zohary 1961). These xeric features include shedding and replacement with smaller leaves, photosynthesizing and water storing parenchyma in the cortical region of the stem and axis (Ginzburg 1963). Interxylary cork rings are developed which have the adaptive value for reducing water loss and restrict the upward passage of water through a narrow zone of the functional secondary xylem (Moss 1940). The present investigation envisages a new record of a peculiar xeromorphic structure in *Artemisia scoparia (Compositae)*.

These special structures in the form of globose to sub-globose bodies (diameter 0.4—1.5 mm) with reddish tip in the early stage and green, gray and finally yellowish at maturity stage, arise singly or in a group (5—10), mostly in the axil or below the leaf on the main stem, as well as on the branches (Fig. 1) and appear from June to October. On account of the position of the structures in the axil of the leaves, these can be regarded as modified branches. This has been confirmed by the endarch condition of the xylem in the transverse section (Fig. 3). Such a modification might favour the plant for moisture conservation in the tissues by terminating the emergence of the lateral branch, thus reducing the exposed surface area to the plant.

Fig. 1. Presence of globose to sub-globose bodies singly and in a group in axil or below leaves on main stem as well as on branches

Anatomical studies on these structures reveal the existence of unbranched, unicellular trichomes arising from the epidermis, and anomocytic type of sunken stomata (Fig. 2) in the epidermis (4—8 stomata/mm^2). The epidermis forming the outermost layer, is uni- to bilayered consisting of compactly arranged, more or less rectangular in (T. S.) parenchymatous cells interrupted at places with stomata. Below the epidermis there is a chlorenchymatous cortex having 2—3 layers of palisade cells with abundant discoid chloroplast. In the initial stages, the entire cortex comprises oval and irregular chlorenchymatous cells with large intercellular spaces. With advancement in age, there develops a row of 4—6 layers of water storing oval parenchymatous cells showing a discrete presence of chloroplast. Branches of vascular bundles have also been observed in the cortex in the initial stages of development of this structure (Fig. 4). Factors like intense illumination and moisture deficit in the soil in arid regions elicit similar modifications in xeric plants (Schields 1950) for reduction in water loss and facilitated gaseous exchange through stomata (Oppenheimer 1960).

In the initial stages the well developed parenchymatous pith covering the central region of the structure is marked. The lignification of the parenchymatous cells first starts at the peripheral portion of the pith. This is followed by the dissolution of the parenchymatous pith from the centre and leads to the development of a normal cavity (Fig. 4). However, the pith at the apical and basal regions of the structure, is intact even at maturity. The pith cavity is associated with 4—5 layers of lignified tissue, some cells of which are elongated and concentrically arranged indicating that the development of thick walls in parenchymatous cells and the additional incorporation of sclerenchyma is a specific phenomenon in a plant experiencing moisture stress (Schneider 1936).

Vascular bundles embedded in sclerenchymatous cells in a ring are collateral with feebly developed xylem and pholem. Such features as the larger cortex accompanied by the

Fig. 2. Anomocytic stomata in the epidermis of the structure. ×1500

Fig. 3. Gross section of the globular structure showing endarch condition of xylam establishing this structure as the modified form of the stem. ×400

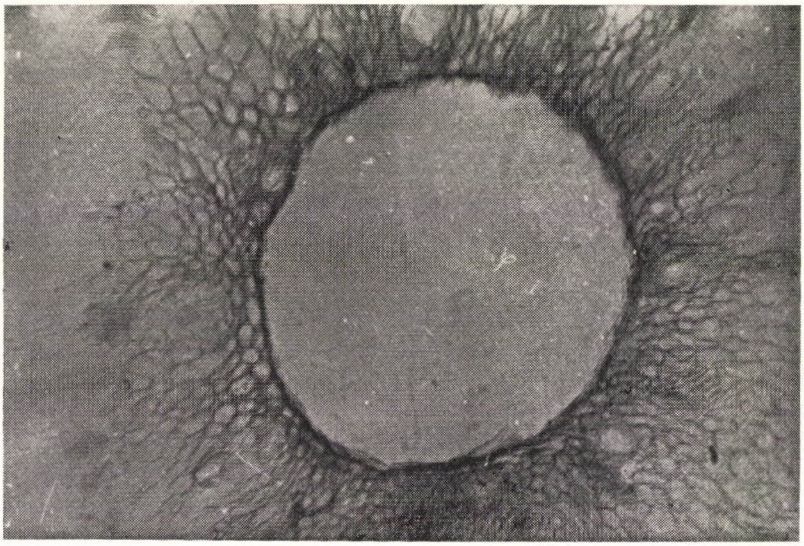

Fig. 4. Cross section of fully developed structure showing the lignified tissue around the central cavity and vascular traces in the cortex. × 180

"Contraction" of the vascular strands around the pith, may be an adaptation by which the vascular tissues are protected from drought in the early stages of development.

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CURRANT HARVESTING MADE EASIER BY SPRAYING WITH ETHREL

Cultivation of certain fruit species is restricted by the high requirement of manual work. This applies especially to the small fruits, e.g. currants; among the tasks of their cultivation the elaboration and introduction of mechanical harvesting methods seem to be the most urgent.

In this field experiments carried on with high frequency shaking machines are very promising (LIEBSTER—SCHIMMELPFENG—BÖHM 1964, KRONENBERG 1964). Harvesting by shaking can only be solved economically, however, with varieties where fruits ripen at the same time, otherwise more or less of the fruits are left on the bushes.

Varieties fulfilling these important requirements should thus be developed or selected. It is another question, however, whether varieties suitable for mechanical harvesting meet other economically important requirements or not.

Beside the aspects of breeding, the problem can be approached from an other side as well. It is known, that the ripening of fruits and the development of the separating layer which causes fruit dropping are in close connection with the intensity of ethylene formation in the plant tissues (BURG—BURG 1965). There is constantly increasing evidence that the processes mentioned are induced by the increased level of ethylene (BURG—BURG 1962, 1966).

Thus it is quite obvious that etylene can be used to control fruit ripening and simultaneously promote the possibility of mechanical harvesting. Owing to the gaseous state of ethylene its direct application in the field encounters serious difficulties, therefore attention is called to materials which, having entered the tissues, either promote a natural ethylene formation or decompose while releasing ethylene. The former group includes the auxins (indole acetic acid, 2.4-D, etc.), while among those belonging to the latter β-hydroxy-ethyl-hydrazine (BOH) and 2-chloroethylphosphonic acid (CEPA) — by its commercial name: Ethrel — should be mentioned. Ethrel preparations produced by the American firm: Amchem Products Inc. have become especially popular in agricultural practice due to their manysided applicability. Ethrel has been tested for the last two years with various fruit species including black currant. In an experiment carried out with the black currant variety Baldwin LUCKWILL (1968) found the solution of 500 ppm concentration to be the most economical. Four days after spraying 86 percent of the fruits could be shaken off by hand compared to 3 percent of the control. Higher concentrations (2000, 5000 ppm) resulted in a premature defoliation as well as in the death of some apical buds and consequently shoot branching three weeks after spraying. Wetting agents increased the efficiency of the chemical (INGRAM 1969).

The first experiments of this kind were carried out at our Institute in the summer of 1970. The necessary spray (Amchem 68—250, one of the Ethrel preparations) was placed at our disposal by the Amchem Products Inc. (Ambler PA., U.S.A.) free of charge. Solutions of 240 and 480 ppm respectively were applied to the following varieties: Black Boscoop and Blacksmith black currants, Jonkheer van Tets and "Hosszúfürtű piros" red currants.

Black currants used in the experiment were five-year-old plants with 25—35 cm high trunks grafted to *Ribes aureum* stocks, while the red currants were six-year-old traditional bushes.

The above concentrations refer to pure active agent. The solutions were adjusted with N/10 NaOH to pH 5.0. The control of the pH was necessary from two points of view. First: the original solution may cause scorching due to its high acidity; second: the amount of ethylene released is in relation with the pH value of the spray. The higher the pH, the more intensive is the ethylene formation (EDGERTON—BLANPIED 1968).

The treatments were performed at the end of June and beginning of July respectively, always on clear warm days of 28—30 °C temperature. At the time of spraying about 60 percent of the fruits in the variety Black Boscoop Giant and 80 percent of those in the variety Blacksmith were black. At the same time the variety Jonkheer van Tets was sprayed at the stage of full ripeness, while in the variety "Hosszúfürtű piros" some less ripe fruits could still be found at the time of the treatment. Spraying was carried out with a common portable sprayer. With the black currants about 1 litre, while with the red currants 2.5 litre solution per bush was used. 2—3 black- and 1 red currant bushes were sprayed per variety and treatment.

Black currant variety Black Boscoop Giant. The fruits began to drop from the bushes as soon as the fourth day after spraying. The fruits dropped were picked up and weighed daily, then harvesting by shaking the bushes was started on the seventh day. Since the plants used in the experiment had trunks, harvesting was carried out by shaking the trunks for 30 seconds. The fruits left on the bushes were picked by hand. In Table 1 the amount of fruits harvested by shaking and those fallen off earlier is expressed as a percentage of the total amount of fruits.

Table 1

The effect of Ethrel spraying on the dropping and hand shakability of fruits in the black currant variety Black Boscoop Giant

Treatment	Percentage of fruits dropped + shaken off
Control	6.8
Ethrel 240 ppm	65.2
Ethrel 480 ppm.	98.5
S.d. 5%	37.6

The data clearly show that spraying with Ethrel was efficient. Treatment with a solution of 480 ppm concentration loosened the separating layer to such an extent that practically all the fruits could be readily shaken off the bushes. At the same time only an insignificant amount of fruit could be harvested from the control plants with this method.

Black currant variety Blacksmith. The fruits began to drop as soon as the third day after the treatment. Harvesting was started on the fourth day. Harvesting operations were carried out by the method used with the variety Black Boscoop Giant.

Spraying with Ethrel successfully promoted the separation of the fruits from the plants with the variety Blacksmith as well (Figs 1, 2). The difference between the control and the treated plants is even more expressed than in the case of the variety Black Boscoop Giant. (Tabelle 2). The fruits dropped without strigs in both varieties.

Table 2

The effect of spraying with Ethrel on the dropping and hand shakability of fruits in the black currant variety Blacksmith

Treatment	Percentage of fruits dropped + shaken off
Control	1.6
Ethrel 480 ppm	98.7
S.d. 5%	2.7

Fig. 1. Untreated Blacksmith black currant variety after shaken

Fig. 2. Blacksmith black currant variety sprayed with Ethrel solution of 480 ppm after shaken

Red currant varieties. The bushes sprayed with Ethrel began to drop their fruits on the fourth day after the treatment. Harvesting began on the fifth day with the variety Jonkheer van Tets, while — for technical reasons — only on the ninth day in the case of the "Hosszúfürtű piros". Each shoot was shaken for 10 seconds. The fruits dropped were graded into two groups, berries with strigs and without, respectively, and were weighed separately. The fruits left on the bushes were picked by hand. The results were expressed as a percentage of the total amount of yield (Table 3).

Table 3

The effect of spraying with Ethrel on dropping and shakability by hand of fruits in the red currant varieties

Variety	Treatment	Percentage proportion of fruits dropped and shaken off to the total amount of yield			Without strigs per With strigs
		Total	Without strigs	With strigs	
Jonkheer	Control	56.1	21.7	34.4	0.63
	Ethrel 240 ppm	82.1	54.6	27.5	1.99
	Ethrel 480 ppm	98.9	87.6	11.3	7.73
"Hosszúfürtű piros"	Control	12.2	11.4	0.8	13.21
	Ethrel 240 ppm	83.5	75.8	7.7	9.88
	Ethrel 480 ppm	93.0	83.5	9.5	8.80

The Ethrel treatment promoted the separation of the berries from the plants in the red currant varieties as well. Bushes sprayed with a solution of 480 ppm dropped practically all fruits when shaken by hand (Fig. 3, 4). In contrast with the black currant varieties a part of the berries dropped with their strigs on. In the variety Jonkheer van Tets the proportion of the fruits that dropped without strigs to those that dropped with their strigs on decidedly grew with an increased concentration of Ethrel, while with the red currant variety "Hosszúfürtű piros" the opposite tendency was observed, though the differences were not significant (Table 3, Fig. 5).

On some shoots the leaves turned yellow under the influence of Ethrel treatments. However, this phenomenon could no longer be observed at the end of August. At the time of harvesting a yellow colour also appeared on the floral axes.

Although the experiments are far from being finished, Ethrel treatments already seem to be suitable for facilitating the harvesting operations of currants. If an appropriate form of bush is developed, the above mentioned treatment makes the full mechanization of fruit harvesting possible. From the point of view of simplifying the operation of shaking, the high cultivation method proved to be ideal (Fig. 6). It is a different problem what the regenerating ability of the plants will be like if branching takes place high above the ground, and whether the costs of producing grafts will not render production too expensive.

The application of Ethrel demands quick harvesting carried out in time, otherwise a considerable proportion of the fruits drops, before the harvest. Thus, the labour capacity of harvesting should be taken in consideration by all means when the size of the area to be sprayed at a time is determined. The exact time when harvesting must by all means be carried out

Fig. 3. Untreated Jonkheer van Tets red currant variety after shaken

Fig. 4. Jonkheer van Tets red currant variety sprayed with Ethrel solution of 480 ppm after shaken

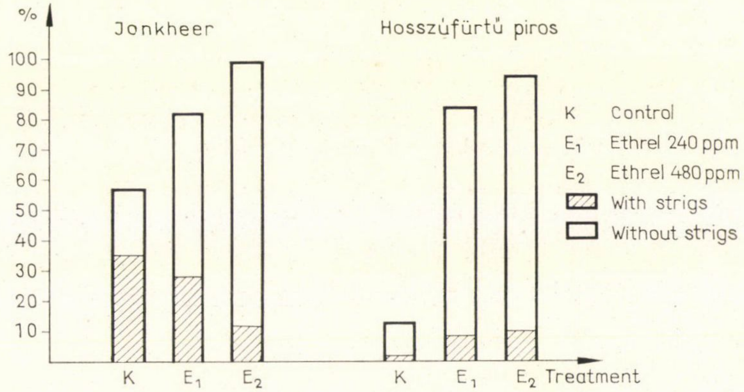

Fig. 5. Effect of spraying with Ethrel on the red currant varieties. Percentage proportion of fruits dropped and shaken off to the total amount of yield

Fig. 6. Five-year-old Black Boscoop Giant black currant plant grafted to Ribes aureum Golden C rootstock

after the treatment depends — naturally — on the ripeness of the fruits and the weather as well.

The question of residue has not been cleared either, so the application of Ethrel is restricted for the time being to experimental treatments. Although as far as it is known, Ethrel decomposes in the plant while free ethylene gas, chloride- and phosphate ions are released, still — as all new chemicals — Ethrel should be used carefully.

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SEASONAL CHANGES OF ASSIMILATING SURFACE AND CHLOROPHYLL CONTENT IN FESTUCETUM VAGINATAE AND SECALETUM CULTUM COMMUNITIES

Light penetrates the leaf surface of plants and enters the chloroplasts and within them the photosynthetic apparatus, where the chlorophyll molecules utilize the light energy in building up organic matters. The light energy uptake of plants is not totally independent of the size of the absorbing surface. Thus, from the point of view of plants and plant communities the assimilating surface and chlorophyll content per m^2 area of the community are of great importance. According to Golley (1965) these parts belong to the structural elements, while production to the function of the community.

Many authors might be listed who dealt with the chlorophyll content of plant communities. Most of them studied the relation of chlorophyll content to production in native and managed communities. Just to mention a few of them: Bray (1960, 1962), Golley (1965), Bliss (1966), Pilát (1967) studied native communities while, Brougham (1960), Sesták (1966) investigated the relation of chlorophyll content to production in cultivated plants. Ovington—Lawrence (1967) studied the changes of chlorophyll content during the vegetative period in four ecosystems. ODUM *et al.* (1958) explained the role of chlorophyll in the process of production under natural conditions. MEDINA—LIETH (1964) examined the relation of chlorophyll content and assimilating surface to production in native and managed communities.

Relationship between chlorophyll content and production — though studied by many — has not been perfectly disclosed so far, therefore the study of the chlorophyll content is a timely subject even today, especially in those series of investigations which include production studies.

In our investigations the changes of chlorophyll content and assimilating surface during the vegetative period were studied in two plant communities *(Festucetum vaginatae danubiale* and *Secaletum cultum)* pointing out the differences between the two communities growing under similar ecological conditions (cf. Simon—Orbán 1972).

Simultaneously with the sample taking (1969), production studies were carried out in rye, the results of which could be compared with data on the chlorophyll content.

The two communities can be characterized by the results obtained and the relationship between the two structural elements of the community examined. What changes are shown in the chlorophyll content and assimilating surface during the vegetative period?

These are the data with which we should like to contribute to the study of the two ecosystems; data characteristic of the communities and utilizable in further investigations.

Description of the site. The Nature Conservation Area is located about 44 km southeast of Budapest. The upper layer of its sandy soil contains calcium carbonate. The former drift-sand has already been bound by the poplars and junipers found on the area. The Natural Conservation Area is a good example of the succession of vegetation on a calciferous sandsoil. Communities developing during the succession of calciferous sand can be found side by side in a relatively small area. From the annual grass of *Brometum tectorum* to the community of lily of the valley and oak *(Convallario-Quercetum)* all stages of succession can be found.

From the above a calciphilous perennial grassland community *(Festucetum vaginatae danubiale)* was chosen. This community is found on sand-hill tops or at dry places between sand-hills. *Festuca vaginata* prefers places where ground water is at a depth of more than 2 metres, and the value of pH is between 7.3 and 8.4 (Soó 1965). Plant species growing on the selected sample plot are presented in Table 1.

Of these species *Festuca vaginata* occurred in the largest numbers on the sample plot. The other species, *Koeleria glauca* — though also growing in large masses — was considerably less than *Festuca vaginata.* In the sample square *Bromus tectorum* and *Secale silvestre* were found in the spring vegetation, while *Potentilla arenaria, Galium verum, Euphorbia segueriana* and *Dianthus serotinus* during the summer. Other plant species occurred but sporadically in the sample square.

The other selected plant community was a rye *(Secaletum cultum)* stand. The rye stand was at a distance of about 2 kms from the perennial grass community, that is sufficiently close for considering the meteorological conditions (precipitation, temperature) identical. The rye was sown likewise into a sandsoil, however, there was an essential difference between the two sites in the water table. The water table of the rye stand was high (about 1 m). Since the meteorological conditions are identical, comparison can be made between the two communities.

Sampling. We took a sample of each of the *Festucetum vaginatae* grassland and the rye stand every month in 1969 by cutting the plants on a quadratic spot of 1 m^2. In the grass community this spot had to be chosen in such a way as to obtain a cover characteristic of the whole community. With rye this did not cause any problem since the cover was rather uniform as a result of drilling. In the case of the *Festucetum vaginatae* community no sample was taken of the moss-lichen level, while in the rye stand the weed-plants were not taken into consideration as their quantity was negligible compared to the mass of rye.

Grass- and rye samples cut were placed in plastic bags and delivered to a laboratory for further processing. In the laboratory the dry and completely withered parts were separated

Table 1

List of plants ocourring from early spring to late autumn stage on the sample spot

Festucetum vaginatae danubiale community

Festuca vaginata
Koeleria glauca
Secale silvestre
Bromus tectorum
Potentilla arenaria
Eringium campestre
Peucedanum arenarium
Onosma arenaria
Galium verum
Euphorbia segueriana
Erysimum diffusum
Silene otites
Myosotis stricta
Medicago minima
Seseli annuum
Asperula cynanchyca
Helichrysum arenarium

Ephedra distachya
Sedum hillebrandii
Astragalus varius
Alyssum tortuosum
Syrenia cana
Echinops ruthenicus
Centaurea arenaria ssp. *tauscheri*
Tragopogon floccosus
Egipactis atrorubens var. *borbásii*
Dianthus diutinus
Linum hirsutum ssp. *glabrescens*
Equisetum ramosissimum
Trigonella monspeliaca
Alcanna tinctorica
Alyssum montanum ssp. *gmelini*
Dianthus serotinus
Colochicum arenarium

from those containing chlorophyll. Then the fresh weight of samples obtained from 1 m^2 each was measured. From this fresh plant material aliquot samples of 1 g per each were taken for determining the chlorophyll content and assimilating surface.

Since in the grass community various kinds of plants occurred in the 1 m^2 sample, we endeavoured to take the 1 g samples proportionally to the weights of the different grasses.

For chlorophyll determination the samples were processed within 24 hours in order to prevent the chlorophyll from a higher degree of decomposition. For the determination of the assimilating surface the samples were pressed and put aside until further processing.

Determination of chlorophyll content. The determination of chlorophyll content was carried out according to the method described by Koski—French (1951) and French (1960) with the aliquot samples used. 1 g samples were homogenized with some quartz in acetone containing $MgCO_3$, and the pigments extracted with toluol. $MgCO_3$ neutralizes the plant acids released.

The solution obtained was centrifuged in order to sediment the floating particles. The completely pure solution was diluted to 20 ml, but no column chromatography was applied as in the original method described by Koski—French (1951) and French (1960), instead the solution obtained was diluted and spectrophotometrically treated. Determination was carried out with a Spektromom 202 spectrophotometer. Chlorophyll "a" was measured at 663 nm, while. chlorophyll "b" at 645 nm, since their maximum light absorption falls within the above spectral ranges. By means of extinction values measured in the spectrophotometer, if the molar extinction coefficient and mole weight are known, the amount of chlorophyll "a + b" can be calculated. Calculations were made with the method of Smith—Benitez (1955) too, and the results were the same. Since the absorption maxima of the two chlorophylls are close to one another in the spectrum, their interaction was adjusted.

From the extinction values obtained the chlorophyll "a + b" content of the 1 g samples was computed. The chlorophyll a + b contents of 1 m^2 sample spots were obtained for *Festucetum vaginatae* and rye by multiplying the fresh weight with this value. Determinations were repeated with 3—5 samples, their mean value gave the results.

Determination of assimilating surfaces. By the term: assimilating surface all parts containing chlorophyll are implied. Thus, it differs from both the leaf area index (LAI) and the leaf area ratio (LAR), being in every case larger than either of them. (The leaf area index is the leaf area per unit land area, m^2/m^2, while the leaf area ratio is the leaf area per unit biomass, dm^2/g, which are very intensely examined especially in cultivated plants.) Assimilating surface determinations were carried out by Medina—Lieth (1964) in cultivated plants and plant communities.

Many methods have been developed so far for surface determination, of which two interesting ones should be mentioned here: Medina and Lieth who also measured the assimilating surface used a light planimeter, while Kemp (1960) elaborated a mathematical method of determining the surface of grasses: he calculated from their length and width.

In the course of our investigations a photographic method was used in the case of samples from both communities. Plant parts contained in the 1 g samples were placed on photo paper in such a way as to prevent overlapping, and illuminated. Having been developed and dried, the pictures of the individual plants were cut out of the paper and weighed with a torsion balance. At the same time pieces of known size were cut out of the photo paper and weighed with the torsion balance. In this way the surface of the individual plant parts could be calculated by means of a simple proportion. The surface values obtained were totalled, the flat parts multiplied by 2 and cylindrical parts by 3.14. Through this calculation the surfaces of the 1 g aliquot samples were obtained. To obtain the assimilating surface of the plant material contained in the 1 m^2 sample, the value obtained for 1 g was multiplied by the fresh weight of the plant material contained in 1 m^2.

Chlorophyll "a + b" content in the communities studied. Data on the chlorophyll content per unit surface of the plant communities are summed up in Table 2. A comparison between the two communities shows great differences in the values. This agrees with the observation of Bray (1960) who found as many as twentyfold differences between the communities studied.

The chlorophyll content determined for a land area unit of 1 m^2 depends on the density and height of the plant stand, Bray (1960) found a close positive correlation between chlorophyll content and plant height, the extent of shading, age of plants (Odum 1959), etc. With all this taken into consideration it is evident that there are fundamental differences between the two communities. The density of the rye stand was much higher than that of the natural perennial grass community where 40—50 per cent of the sand surface was uncovered. There were considerable differences in the height of plants too, since the rye grows more than one metre high during the vegetative period, while the grass community hardly reaches half a meter of height.

At the time the two communities attain maximum chlorophyll content, the difference between them is 2.6-fold.

When plotting the data (Fig. 1 and 2), the change of the chlorophyll content during the vegetative period becomes conspicuous. A similar phenomenon was observed by Ovington—Lawrence (1967) in prairie-, savanna-, maize field- and oak-forest ecosystems.

Our measurements gave maximum curves, in both cases where the maxima were in different parts of the vegetative period. In the case of rye the maximum value was in May (in 1969 it was 1178 mg/m^2). The increase before and decrease after the maximum was very

Table 2

Data on chlorophyll "a + b", assimilating surface and production in samples taken once a month

	1	2	3	4	5	6	7	8	9
Secaletum cultum	IV	157.132	1.75	0.011	0.92	37.50	50.00	902.75	952.75
	V	1178.010	3.59	0.032	1.50	235.00	305.00	1089.00	1394.00
	VI	465.896	1.87	0.024	1.32	461.50	512.72	630.75	1143.50
	VII	—	—	—	—	61.00	682.50	501.00	1183.50
	VIII	—	—	—	—	20.00	25.00	515.00	540.00
						phytomass data of 1968			
Festucetum vaginatae	IV	34.508	0.185	0.020	1.14	50.375	77.625	1634.125	1711.750
	V	290.156	0.538	0.052	3.02	68.875	170.000	2527.125	2697.125
	VI	438.675	0.933	0.044	2.90	32.875	175.875	2180.250	2356.125
	VII	—	—	—	—	48.750	179.625	1630.750	1810.375
	VIII	241.682	0.651	0.036	2.91	32.125	109.250	970.875	1080.125
	IX	182.233	0.391	0.044	3.10	97.500	224.125	1159.500	1383.625

The data of the table: (1) months, (2) chlorophyll „a + b" mg/m^2, (3) assimilating surface m^2/m^2, (4) chlorophyll/assimilating surface mg/cm^2, (5) chlorophyll concentration mg/g, (6) dry weight of above-ground plant material g/m^2, (7) dry weight of above-ground living + dead material g/m^2, (8) dry weight of root g/m^2, (9) dry weight of total material g/m^2. Production data of the grass community in 1968.

rapid. The rapid increase can be attributed to the fact that in April—May rye displays a sudden vertical elongation, develops its fertile shoot. This sudden growth involves an increase in the assimilating surface and the above-ground biomass. After the maximum has been reached a rapid decrease can be observed in the chlorophyll content. This is caused by the fact that by the end of June the rye becomes mature, and as the plant parts become successively yellow, the chlorophyll content gradually decreases. When extending the curve we can see that by the end of June the chlorophyll content reaches zero.

With the open perennial grass community the maximum appears a month later, its value is 438 mg/m^2. In this case the situation is more complicated, as here more than one kind of plants occur on the 1 m^2 sample spot, which have different chlorophyll contents; one species contains more, the other less chlorophyll. These values appear totalled, and the maxima obtained are not as high as with the rye stand, because the chlorophyll content of plants which are in different phases of development is lost during the sampling, so the values are equalized. E.g.: During the vegetative period beside the perennial *Festuca vaginata* and *Koeleria glauca* — which are present throughout the whole year — other species appear in different periods. The result is shown in the shape of the curve, as the rapid increase of chlorophyll content in spring is probably caused by the presence of annual plants. After the maximum the change is presumably determined by the *Festuca vaginata*, which occurs in the largest masses in this place and reaches full development by this time. It is probable that the chlorophyll content in the *Festucetum vaginatae* community never falls to zero, but there are likely to be great differences in the activity of the chlorophyll.

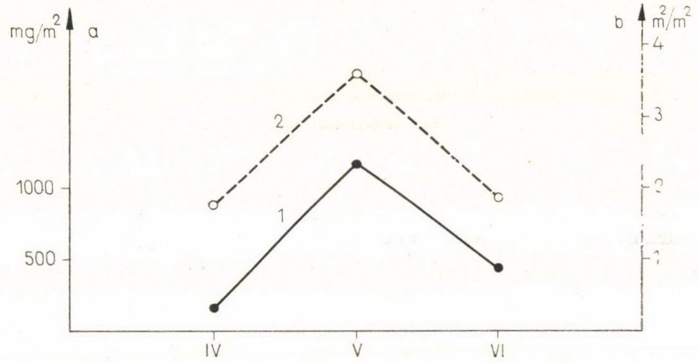

Fig. 1. Changes in the chlorophyll content (1) and assimilating surface (2) of rye stand during the period of investigation. Abscissa: months, "a" ordinate: chlorophyll "a + b" content mg/m², "b" ordinate: assimilating surface m²/m²

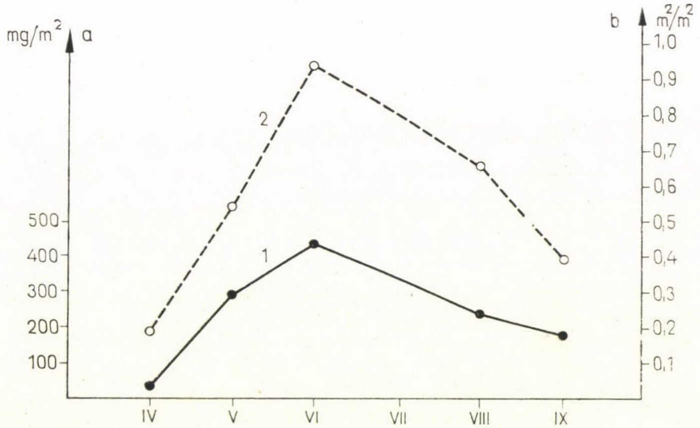

Fig. 2. Changes in the chlorophyll content (1) and assimilating surface (2) of *Festucetum vaginatae* community during the period of investigation. Abscissa: months, "a" ordinate: chlorophyll "a + b" content mg/m², "b" ordinate: assimilating surface m²/m²

We calculated the chlorophyll concentration for 1 g leaf. The changes shown by this value during the vegetative period differ from those shown by the chlorophyll content per 1 m² area. At the beginning of the vegetative period, in April—May, the chlorophyll content in 1 g leaf increases considerably. In the case of *Festucetum vaginatae* the chlorophyll content in 1 g leaf at the time it reaches maximum in May is twice as high as in the rye stand, which has its maximum in the same month. After the maximum less changes occur in the chlorophyll concentration. The data on the chlorophyll concentration are shown in Table 2 while its monthly changes are shown in Fig. 4.

Changes in the assimilating surface. The monthly changes in the assimilating surface are shown in Fig. 1 and 2, while their data are shown in Table 2.

Similarly to the chlorophyll content, here too, there are great differences between the two communities. The assimilating surface too — like the chlorophyll content — probably varies from species to species, and thus the assimilating surfaces of the communities are possibly also different and characteristic of the community.

When comparing the maxima we find that the value of the rye stand is 3.5 m^2/m^2, while that of the *Festucetum vaginatae* community is not even as high as 1 m^2/m^2 when reaching maximum in June.

Although so far authors have referred to the leaf area index when suggesting that changes in the leaf surface depend on the type of plant, stand density, water- and nutrient supply (P, K, N) and light (DONALD 1963, BLACKMAN 1968, NICHIPOROVICH 1968), we think that this statement is true in the case of the assimilating surface too, although investigations in this respect have not been made. It can be clearly seen, however, that the assimilating surfaces of plant species are different and also change in time. In our investigations the assimilating surface of the rye stand was found to be much larger than that of the less dense perennial sand grassland. The rye stand reaches its maximum assimilating surface in May (in 1969 3.5 m^2/m^2 was measured at that time). From the values of the samples taken in April to the maximum the increase is rapid; from the value of 1.75 m^2/m^2 the assimilating surface increases twice as much. The increase of the assimilating surface can be explained in the same way as that of the chlorophyll content.

In the case of *Festucetum vaginatae* communities the maximum value can be found in samples taken in June, thus the difference between the two ecosystems is shown here too. The maximum assimilating surface of the grassland was 0.933 m^2/m^2, 3.7-times less than in the managed community.

Chlorophyll "a + b" content per 1 cm² assimilating surface. In the course of autoecological studies this value is used for characterizing the plant species chosen (WHITTAKER—GARFINE 1962).

It is worth examining how this value changes in the different plant communities. If the chlorophyll contents determined monthly are divided by the assimilating surface (cm^2), the chlorophyll content belonging to 1 cm^2 assimilating surface is obtained. This value is the distribution of the chlorophyll content over the assimilating surface of the community. Of course, this does not mean a two-dimension distribution, but the amount of chlorophyll related to 1 cm^2 assimilating surface of the community. Thus, the ratio of chlorophyll/assimilating surface is characteristic of the whole plant community, a value formed by the joint effect of the different plants in the community and by the ecological factors influencing the given community.

The ratio of chlorophyll/assimilating surface shows changes during the vegetative period (Table 2, Fig. 3). When comparing the values obtained in the case of the rye stand on the one hand, and *Festucetum vaginatae* communities on the other, we find that on the large assimilating surface of the rye stand the chlorophyll pigments are distributed on a larger area. Chlorophyll content per 1 cm^2 ranges from 0.011 to 0.032 mg in the rye stand and from 0.020 to 0.052 mg in the grass community during the vegetative period. These changes in the chlorophyll content and assimilating surface are caused by changes occurring during the vegetative period.

Relationship between chlorophyll content and assimilating surface. We tried to find an adequate correlation between the two characteristic structural values of the two communities. The correlation coefficient between the values of chlorophyll content and assimilating surface is +0.90, which is a very high value. This value means that there is a linear correlation between the chlorophyll content and the assimilating surface: the chlorophyll content changes to the same degree as the assimilating surface. The data of both communities were used simultaneously when calculating the correlation. The correlation calculations were carried out on the basis of the method of SVÁB (1967).

Production results. The production data of the communities we studied were determined by Kovács—Láng—Szabó (1971). In the *Festucetum vaginatae* community the production studies were carried out in 1968, in the rye stand the samples for measuring the phytomass were taken simultaneously with the chlorophyll sampling, in 1969. The phytomass data of the two communities are shown in Table 2. The *Festucetum vaginatae* data are only of informative character, as the samples were taken a year earlier. In the case of the rye stand a comparison can be made between the chlorophyll content and production.

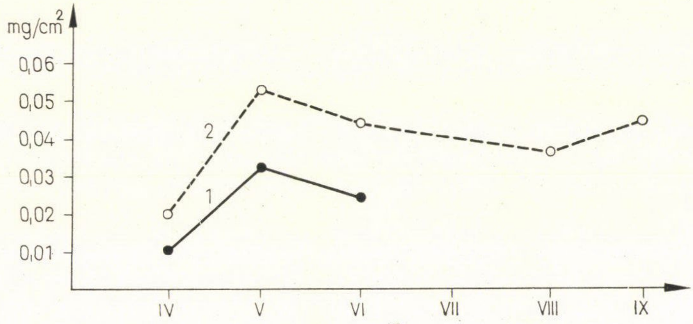

Fig. 3. Changes in the ratio of chlorophyll to assimilating surface during the period of investigation in the case of rye stand (1) and *Festucetum vaginatae* community (2). Abscissa: months, ordinate: chlorophyll/assimilating surface mg/cm²

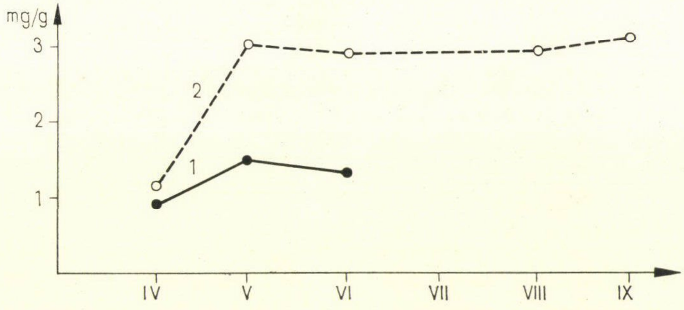

Fig. 4. Changes in the chlorophyll concentration during the period of investigation in the case of rye stand (1) and *Festucetum vaginatae* community (2). Abscisse: months, ordinate: chlorophyll concentration mg/g

Many researchers have dealt with the chlorophyll content of communities. It has been pointed out that the chlorophyll content is different in the various communities and increases with the age of the succession stage of the communities from meadows to forests (Bray 1960).

From the data of all the communities examined the range of values within which chlorophyll content per 1 m² land area varies in the different plant communities was determined. According to the values determined for chlorophyll "a" the chlorophyll content per 1 m² ranges between 0.1 and 13.3 g/m²; in native terrestrial and aquatic communities (Odum *et al.* 1958) 0.1—3.0 g/m²; at more productive sites, in a Typha marsh Bray (1960) obtained a value of 4.6 g/m², while with Populus tremuloides 5.9 g/m²; **Aruga—Monsi** (1963) found the highest value in the case of evergreen gallery forests: 13.3 g/m². Our own data agree with the above range of values.

Most researchers examined the chlorophyll content from the point of view of its relationship to the production of the plant or of the community. **Bray** (1960, 1962) pointed out a close correlation between chlorophyll "a + b" content and above-ground dry weight in herbaceous vegetations and suggested using the chlorophyll content as an index in production calculations. This method has already been used by hydrobotanists, and **Odum** (1959) considered it useful in certain cases of terrestrial ecosystems too. Later correlations turned out to be not so close in every case (e.g. **Bliss** 1966). Others pointed out that the correlation between chlorophyll and production existed only in certain part of the vegetative period, when the ecological factors are optimal for the plant community (**Pilát** 1967).

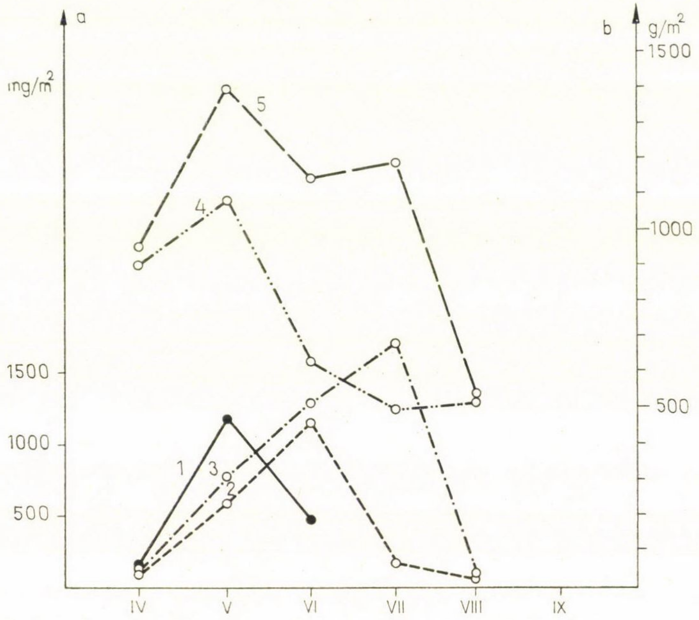

Fig. 5. Changes of phytomass and chlorophyll content in a rye stand during the period of investigation. (1) chlorophyll content, (2) above-ground living phytomass, (3) above-ground total phytomass, (4) root, (5) total phytomass. Abscissa: months, "a" ordinate: chlorophyll content mg/m², "b" ordinate: phytomass dry weight g/m²

When studying the chlorophyll- and production data of the rye stand we can see that at the beginning of the vegetative period production increases parallel with the increase in the chlorophyll content (Fig. 5). In our opinion it is in this period that the relationship between production and chlorophyll content should be studied by taking samples at shorter intervals.

Assimilating surfaces were determined by **Medina—Lieth** (1964) in cultivated plants and in two plant communities. They found no correlation between assimilating surface and production, except when dividing the chlorophyll content (in g) by the assimilating surface (in m²). Thus, the assimilating surface shows different trends from the leaf area index, and we think it expresses the relation of chlorophyll content and surface all the more.

It should be mentioned that the relation of leaf surface to production depends on how much the leaf surface can utilize of the light received. This depends on the position of the leaf and the incidence of the sunshine. This may be true in the case of the assimilating surface too. Several authors use a correction when calculating the useful surface (BROUGHAM 1960, GOLLEY 1965, KUROIWA 1968). This correction may really be valuable when daily examinations are performed with a single plant or with the population of a species, or when artificial illumination is employed. When, however, investigations are carried on by monthly sample taking, and plant communities are considered, the use of this correction has — in our opinion — no meaning, especially when cloudy and sunny days alternate on the given area, as e.g. in our case.

To the best of our knowledge the chlorophyll content per 1 cm^2 assimilating surface of plant communities has not been investigated so far. The ratio of chlorophyll/assimilating surface is — in our opinion — characteristic of the whole community, a value formed by the joint effect of all plants contained in the community, and by the totality of ecological factors influencing the community in a given period. It is supposed, thus, that each community has a value which shows changes according to the ecological factors, but is characteristic of the community or ecosystem.

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Prepared at the Botanical Department of the Museum of Natural Sciences, Budapest.

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EFFECT OF FORMAMIDE AND DIMETHYL-FORMAMIDE ON GERMINATION

From the direction of the galactic centre, where the Sagittarius configuration lies, besides carbon monoxide- and methyl-alcohol molecules belonging to the interstellar nebulae, formamide too was registered by the West-Virginian (USA) Green Bank National Observatory of Radioastronomy (Föld és Ég, 1971).

It is known that formamide — or formic amide — is an excellent solvent of polar nature, with an increasing industrial importance (FODOR 1960).

From a biological point of view it is an important fact that formamide can be used for the direct synthesis of complicated heterocycles; e.g. the simplest in vitro synthesis of purine, this important basic skeleton of nucleic acids, was also effected from this molecule in a closed system at 200 °C (BREDERECK—ULMER—WALDMANN 1956). (In vivo this process might have taken place in a similar way during the history of the earth!) Xanthene, the basic compound of plant alkaloids that has a purine skeleton, could be synthetized from formamide in a similar way (BREDERECK 1957).

In possession of the above important and generally known facts we tried to find out the extent of the phytotoxic action of formamide by germination tests well-proved in experiments carried out so far. Beside formamide we also examined some structural analogues such as dimethyl-formamide (a solvent able to dissolve macromolecules), formaldehyde, ethylalcohol and — as a control — water.

Beans and wheats of high germinating ability were used as test plants. The seeds were placed in Petri dishes in 4×100 replications and germinated at 20 °C in the dark. The seeds were irrigated with 1, 5 and 10 per cent aqueous solutions of the compounds used. From the analysis of germinating power and -percentage the following conclusions were drawn (Table 1).

The simple structured compounds used are toxic especially at concentrations of 5 and 10 per cent. At such concentrations the germination power decreases and germination is protracted. The germinated seedlings showed no abnormality. Of the solvents generally known to have a high polarity ethylalcohol and formaldehyde are the most toxic. Formamide is not toxic either to mono- or to dicotyledons at a concentration of 1 per cent, and even at a 5 per cent concentration it is only slightly toxic ! However, at a concentration of 10 per cent even this compound renders germination impossible. Dimethyl-formamide, a dimethyl derivative, the structural analogue of formamide is also only slightly toxic.

Table 1

Phytotoxic action of some polar solvent on germination in wheat and bean

Name of the compound	Test plant	Concentration used %	Germination percentage
water	bean		95
	wheat		95
ethylalcohol	bean	1	80
		5	0
	wheat	1	94
		5	63
		10	0
formaldehyde	bean	1	92
		5	0
	wheat	1	20
		5	0
formamide	bean	1	95 !
		5	68 !
		10	0
	wheat	1	95 !
		5	83 !
		10	0
dimethyl-formamide	bean	1	93 !
		5	5
		10	0
	wheat	1	95 !
		5	0

Thus, it was found that the simple structured organic compounds examined were highly toxic at concentrations of 5 and 10 per cent, while the 1 per cent formamide was considered indifferent from a plant physiological point of view. This is an important fact, as formamide can be used to solve thermosensitive compounds not readily dissolved in water, mainly hormones or homone-like substances of high biological effect. It mostly has a chemical indifference too, compounds can be dissolved in it without decomposing.

It is probable that formamide is one of the most essential molecules of biologically important compounds generally occurring in nature.

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SYMBOLS OF VIRUS AND MYCOPLASMA CRYPTOGRAMS*

Many new results have been obtained in the course of investigations on plant viruses and mycoplasmas in the past several years. The rapid increase in the number of viruses and mycoplasmas is especially remarkable. E.g. the publication entitled Plant Virus Names (edited by Martyn 1968) issued by the Commonwealth Mycological Institute, Kew, Surrey (England) in 1968 enlists more than 600 virus names (including mycoplasmas). The first supplement of the publication which contains further viruses and mycoplasmas described between 1968 and 1970 enlists the names of nearly 100 recently described viruses and mycoplasmas (cf. Martyn 1971). The characteristics of the viruses and mycoplasmas described earlier and recently can be memorized with increasing difficulties owing to their large numbers. The decision of the IXth International Congress for Microbiology by which the Cryptogram Subcommittee has been brought into existence is, therefore, especially welcome. The Cryptogram Subcommittee consists of seven members with J. A. Gibbs, Rothamsted Experimental Station, Harpenden, Herts (England) virologist as Chairman. The Cryptogram Subcommittee was aimed at supplementing the existing popular English virus names with cryptograms (for example, R/1 : 2/5 : : E/E : S/* for tobacco mosaic virus), thus giving information on the most essential characteristics of viruses. The cryptograms contain four pairs of symbols representing the following characteristics**:

1st pair. Type of nucleid acid/Strandedness of nucleic acid
Symbols for type of nucleic acid
R = RNA
D = DNA
Symbols for strandedness

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* Authors are requested by the Editorial Office of Acta Agronomica Acad. Sci. Hung. to complete their papers on viruses and mycoplasmas with cryptograms.

** Considering that at present the cryptograms elaborated by Gibbs (1968) and those modified by Lapierre—Spire (1969) are equally in use, we list both Gibbs' and Lapierre—Spire's symbols when explaining the individual symbols.

1 = single-stranded
2 = double-stranded

2nd pair. Molecular weight of nucleic acid (in millions)/Percentage of nucleic acid in infective particles

3rd pair. Outline of particle/Outline of "nucleocapsid" (the nucleic acid plus the protein most closely in contact with it)

Symbols for both properties according to GIBBS (1968)
S = essentially spherical
E = elongated with parallel sides, ends not rounded
U = elongated with parallel sides, end(s) rounded
X = complex or none of above

Symbols for both properties modified according to LAPIERRE—SPIRE (1969)
M = external membrane
U = elongated particles
B = bacilliform particles
S = more or less spherical particles
O = no external membrane
H = helical structure
I = cubic symmetry
MU = external membrane, elongated particles, at least one end rounded
ME = external membrane, elongated particles, ends not rounded
MS = external membrane, spherical particles

4th pair. Kinds of host infected/Kinds of vector

Symbols for kinds of host
A = Actinomycete
B = Bacterium
AB = Blue alga
F = Fungus
I = Invertebrate
S = Seed plant
V = Vertebrate

Symbols for kinds of vector
Ac = Mite and tick *(Acarina, Arachnida)*
Al = White-fly *(Aleyrodidae, Hemiptera, Insecta)*
Ap = Aphid *(Aphididae, Hemiptera, Insecta)*
Au = Leaf-, plant-, or tree-hopper *(Auchenorrhyncha, Hemiptera)*
Cc = Mealy-bug *(Coccidae, Hemiptera)*
Cl = Beetle *(Coleoptera, Insecta)*
Di = Fly and mosquito *(Diptera, Insecta)*
Fu = Fungus *(Chytridiales* and *Plasmodiophorales, Fungi)*
Gy = Mirid, Piesmid, or Tingid bug *(Gymnocerata, Hemiptera)*
N = Nematode *(Nematoda)*
Ps = Psyllid *(Psyllidae, Hemiptera)*
Si = Flea *(Siphonaptera, Insecta)*
Th = Thrips *(Thysanoptera, Insecta)*
Ve = Vectors known but none of above
O = Spread without vectors

Symbols for all pairs
* = Property of the virus is not known
( ) = Enclosed information is doubtful or unconfirmed

Certain properties of mycoplasmas which cause the yellow type diseases discovered some five years ago and are partly similar to, partly different from the viruses, as to some of their properties can also be characterized with cryptograms. Although it must be mentioned here that the cryptogram symbols elaborated for the viruses require certain modifications in the case of the mycoplasmas, first of all in the 1st, 2nd and 3rd pairs. The 4th pair of the cryptograms (kinds of host infected/kinds of vector) are however, suitable for the characterization of mycoplasmas. Properties symbolized in the 1st, 2nd and 3rd cryptogram pairs are hardly —if at all—known in the case of mycoplasmas (see type of nucleic acid in the 1st pair). According to our present knowledge only the properties symbolized in the 4th pair of cryptograms are suitable for the characterization of mycoplasmas (for example, */* : */* : */* : S,I/Au for aster yellows mycoplasma, cf. GIBBS 1968).

Thus, the cryptograms contain the most essential characteristics known so far of viruses and mycoplasmas. Cryptograms containing information are of especially great importance in publications. The information has a significance mainly for those who have not dealt with the virus- or mycoplasma pathogen discussed in the given paper, so their knowledge of the properties of these pathogens is deficient. In scientific papers cryptograms had better be presented in brackets with the first mentioning of viruses or mycoplasmas. E.g.: "For studies on the virus-host relationship two strains of potato virus X (R/1 : */6 : E/E : S/(Fu); cf. GIBBS 1968) were also included." Or "For studies on the virus-host relationship two strains of potato virus X (R/1 : 2.4/6 : O/H : S/(Fu); LAPIERRE—SPIRE 1969) were also included":

Cryptograms of viruses and mycoplasmas can be found in publications by GIBBS (1968, in: MARTYN 1968, 1971), GIBBS (1969), LAPIERRE—SPIRE (1969), ANONYMOUS (1970), GIBBS *et al.* (1970, in: Description of Plant Viruses) and HORVÁTH (1972). The latter publication contains the Hungarian equivalents of the popular English names of viruses and mycoplasmas.

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WINTER BARLEY U 259

Taxonomical place: *Hordeum vulgare* L. convar. *vulgare* Msf. var. *parallellum* Körn. (Mansfeld 1950).

Origin: obtained by crossing the naked barley with the variety Mezőhegyesi 68.

Beginning of breeding: in Hortobágy, continued from 1951 at Törökbálint, then maintained from 1963 at Karcag.

Breeder: Károly Udvaros (deceased).

State qualification: previously certified variety (1963).

General characterization: irregularly six-rowed barley, with good winter-hardiness, early ripening and good productivity.

Morphological description:

Root system: vigorous deep penetrating

Shoot system: with its rapid early development and stooling forms a thick stand. Leaves (shoots) of young plants prostrate in autumn.

Stem: 97.4 cm long on an average (ranging between 73 and 109 cm). Satisfactory stability (value: 4.3; 5 = perfectly stable); nodes cylindrical.

Foliage: broad, short, dark-green, linear-lanceolate leaves.

Ear: irregularly six-rowed, bending, medium long, with closely set grains; aristae of purplish red colour. With a normal sowing ear number per m^2 ranges from 300 to 436.

Caryopsis: large, full; thousand-grain-weight 40—45 g (ranges between 36 and 48 g); hl-weight 63—64 kg. Ratio of ear/straw: 1.2—2. Digestible protein content of grains is 7.5—9.4 per cent.

Biological characters

Germination: cardinal points: minimum 3.2 °C, optimum 20 °C, maximum 35 °C (Mándy 1966).

Vegetation period: from sowing to ripeness 257—274 days (Horváth 1963, Piacsek 1966).

Development: growth vigour good. The earliest Hungarian barley variety. It ripens 3—4 days earlier than the early variety Beta 40. Early development (Mándy 1967).

Winter hardiness: fairly good, similar to the variety Lédeci Beta.

Resistance to diseases: moderately resistant to powdery mildew, resistant to smut (Piacsek 1966).

Demands on farm technology:

Sowing: the optimum time of sowing is the first decade of October (Mándy 1968). Seed requirement- 2—2.5 million germs per cad. hold (1 cad. hold = 1.422 acres).

Soil: it is not particular about soils but loess soils of high nutrient content are favourable.

Productivity: grain yield 14—20 q/cad. hold, straw yield 24—39 q/cad. hold (Horváth 1963, Piacsek 1966).

Growing district: central and northern part of the Great Hungarian Plain.

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FORUM

THE GENETICAL RELATIONS OF PREFORMATION AND EPIGENESIS

History

The idea that the adult characters and shapes of a living organism are already present in the gametes in a smaller form (preformation) arose in the 17th and 18th centuries. The preformationists maintained that during embryonic development this miniature organism would grow to reach full size. The preformation theory had two principal trends. The animalculists (Leeuwenhoek, Hamm, Spallanzani, Hartsacker and Swammerdam) held that the tiny living organism was to be found in the sperm, and that after obtaining nutrient in the ovum due to fertilization it would begin to grow. The ovists (Malpighi, Haller and Leibnitz), however, assumed that the miniature organism was to be found in the egg.

In the middle of the 18th century new ideas sprang up in opposition to the preformationists' views. The epigeneticians (Wolff, Baer and Driesch) denied preformationism. They maintained that the egg cell was an undifferentiated mass without structure from which a differentiated individual possessing a complex structure and various functions would develop in the course of ontogenesis (epigenesis). They also held that there was no predetermination. In contrast to the preformationists' view they thought the appearance of a new feature to be possible.

The idea of preformation is much more unequivocal than that of epigenesis. This might perhaps account for the fact that over the past decades a number of researchers have tried to define the idea of epigenesis.

Definitions

According to WADDINGTON (1960) the "epigenetic system" is "the sequence of causal processes which bring about the development of the fertilized zygote into the adult capable of reproduction". The "epigenetic momentum" has an important role in the processes of ontogenesis. WADDINGTON (1947) illustrates his idea by a ball rolling down in a system of valleys which he describes as "epigenetic landscape".

While analysing developmental processes GOLDSCHMIDT (1955) came to the conclusion that without the idea of "epigenetic momentum" described by Waddington ". . . it is difficult to understand the genetic control of development . . .".

A number of researchers define the term "epigenesis", and "epigenetic" as a formation of a new individual structure guided by an inherited development system (JEPSEN—MAYR—SIMPSON 1949); that is the process of development; or as the interaction of genetic factors during development (MAYR 1963). According to SINNOTT—DUNN—DOBZHANSKY (1958) "epigenesis is the descriptive term applied to the succession of changes by which the organism passes through stages, more or less distinct from each other, in which new parts appear which were not preformed".

The above mentioned definitions of epigenesis are ambiguous because there are no sharp distinctions between the genetic and non-genetic processes of development. A new feature, which has not been preformed, can be formed as a result of mutation. A mutational change, however, can preform the formation of this new feature in further progenies.

NANNEY (1958) distinguishes two types in the cellular control system: one ensuring the replication of the template, in other words, the "genetic system" while the other is a system regulating the expression of specificities and playing an important part in the developmental processes. Nanney terms this as "epigenetic system". Nanney maintains that mutual exclusion and simultaneous expression are two important characteristics of the epigenetic system which can occur not only in the cytoplasm but also in the nucleus.

ELSASSER (1962) does not reduce the biological laws to a special limiting case of physical processes, but he holds that ". . . physical laws may be thought of as a limiting case of organismic laws." "It is convenient to have a term for those aspects of organismic laws that cannot be reduced to pure physics; we shall call these components epigenetic". He uses the term "epigenetic" instead of the former word "biotonic" (1962). Elsasser uses the term "epigenetic" to denote the organismic laws that cannot be deduced from physics.

In ELSASSER's (1958) view embryonic development is chiefly regulated by biotonic laws. Thus morphogenesis, according to the above given definition, is an epigenetic process since these laws cannot be reduced to pure physics. On the basis of the preformationists' concept the information content of the fertilized egg is equal to that of the adult. Here the assumption should, however, be made that the storage of information is complete. Since in living organisms physical predictability is missing, thus the storage of information cannot be complete either (ELSASSER 1962). According to Elsasser's epigenetic view the fertilized egg does not contain the complete information content of an adult individual.

RAVEN (1961) criticizes the introduction of Elsasser's biotonic laws by saying that ". . . it is very difficult to see the difference between Elsasser's 'biotonic laws' and Driesch's Ganzheitskausalität." Raven holds that during morphogenesis the organisms obtain a large amount of irrelevant information from the environments. He maintains that there is a specific information which is conveyed from generation to generation through the gametes. This is present in the zygote as well as in the adult organism. In an adult, however, the redundancy factor is higher than in the egg. If all the factors are considered, the information content of an adult is higher than that of an egg. (This means epigenesis.)

Commentary

Researchers do not use the term of information unequivocally in each case. In a statistical sense the degree of disorder can be expressed quantitatively by the information content. In this sense the degree of organization increases during the ontogenesis; this should bring about a decrease in the information content. If the information content rose it would go beyond the imagination that organization can appear during the ontogenesis.

If the term "information" is meant to be a certain "message" or program, the specific or genetic information must be regarded as a program tape which directs the operation of the system. In this case this "program" is present in each cell of the adult (in somatic cells and zygote, too).

It has been demonstrated that the whole organism can be regenerated from a single somatic cell (BUTENKO—YAKOVLEVA 1962, STEWARD *et al.* 1958).

On the basis of what has been discussed above it can be concluded that the term "preformation" denotes the determination of processes and features while "epigenesis" is used to term non-predetermined processes and features regulated by the momentary changes of external and internal factors.

The phenotype of organisms is the result of the joint influence of genetical and environmental factors. However, the genotype does not determine a concrete phenotype but a certain range of the possibilities of the phenotype concerned. Thus the genotype of organisms determines their reaction spectrum (norm of reaction). But it is the environmental influences that determine which particular phenotypical process should take place within a given reaction spectrum (range of reaction).

So the process of ontogenesis is regulated by two fundamental mechanisms: one is the so-called preformational mechanism, or to use a better term, determinative mechanism which determines ("preforms") the possibilities of the phenotype, that is, the reaction spectrum of organisms. (Nanney's "genetic system"?)

Within the reaction spectrum a concrete phenotype (that is, the actual expression of a certain possibility) is realized by the epigenetic mechanism, or, to use a more exact term, the realizer mechanism. (Nanney's "epigenetic system"?)

The determinative mechanism corresponds to the genetic information (program) coded in the genetic material (e.g. DNA), namely, it is the genotype. The determinative mechanism can be changed by mutation, chromosome aberration, crossing over, position effect, and by the transfer of chromosomes, episomes, DNA (transformation).

The realizer mechanism accomplishes the expression of the genetic program determined by the genetic material. The primary genetic information content does not change if the realizer mechanism is modified (e.g. modification, homeostasis, phenocopy). The realizer mechanism includes replication, transcription, translation, genetic and physiological regulation systems.

The ontogenesis is guided by the determinative mechanism. The operation of the realizer mechanism also depends on the determinative mechanism. Both mechanisms operate simultaneously in the process of growth, differentiation and organization. That is why the process of ontogenesis cannot simply be considered as an "epigenetic" process. Thus "epigenetic" factors cannot serve as decisive arguments for the explanation of an individual's development. All in all we cannot become familiar with the so far unknown factors of morphogenesis if we try to use an obscure idea (epigenesis).

Not only multicellular but unicellular organisms too have a determinative and realizer mechanism. Viruses are an exception which have no complete realizer mechanism but do have a determinative mechanism. (They are not living organisms.) Viruses use the realizer mechanism of the host cells at their multiplication. The combination of the determinative and the realizer mechanism was an important step in the origin of life. There seems to be an outspoken preference for referring to the presently unexplored processes of development, differentiation and ontogenesis as "epigenetic processes". Studying the processes of ontogenesis it is a better basis for research to adopt the terms determinative and realizer mechanisms. In studying ontogenesis, the unknown parts and laws of the processes of determination and realization should be examined. And as far as the term "epigenesis" is concerned, it would be desirable to put this obscure and ambigous word into a museum.

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CONTRIBUTIONS TO THE PAPER OF GY. BORKA, K. BORKA: "DAILY AND ANNUAL RHYTHM OF WATER REGIME INDICES IN PEACH VARIETIES OF VARIOUS RIPENING TIME" PUBLISHED IN THIS PERIODICAL, *21*, (1—2) 243—252.

ARE THE EXPERIMENTAL TECHNIQUES USED BY GY. BORKA AND K. BORKA APPROPRIATE?

My overall impression is that the manuscript of the above paper reads as though it might have been written some ten or more years ago, rather than in 1971. This impression was shaped by a consideration of the nature of many of the experimental techniques used and also of the concepts discussed.

With regard to the techniques used, I found the two references given on the method of transpiration measurement to be inadequate. The first (BORKA 1967) is to a Ph. D. thesis and so is not readily available; the second (PARCEVAUX 1965) is almost entirely devoted to the analysis of an electric analogue of diffusion resistances associated with a leaf, no technique for measuring transpiration being specifically mentioned. However, Parcevaux's paper contains references to contemporary work of his in which "relative transpiration" has been measured by comparing rates of water loss from leaf discs with evaporation rates from a Piche evaporimeter, a correction being applied for any temperature difference between the leaf and the wet paper.

If the present paper has used a similar technique, then I must point out that, initially at least, rates of water loss from excised leaves or from leaf discs are likely to be considerably (approximately 50—100%) greater than the corresponding rates from attached leaves in the same environment. This has been clearly demonstrated by FRANCO—MAGALHAES (1965) and FRANCO—INFORZATO (1967). The effect is brought about by a hydropassive opening of the stomata, probably as a result of faster turgor loss by the surrounding epidermal cells than by the guard cells (FALK 1966). PARCEVAUX (1965) was aware of this source of error, and commented that changes in stomatal aperture following leaf excision are rare in cultivated plants. Unfortunately, a perusual of the literature shows that this is in fact not the case. Thus, transitory stomatal opening following leaf excision has been reported in coffee, runner beans and castor oil (FRANCO—MAGALHAES 1965), in cotton and sweet pepper (BARRS—KLEPPER 1968), in citrus and tomato (ANDERSON *et al.* 1954), and in all except one of thirty species examined by KAMP (1930, cited by FALK 1966). Furthermore, the magnitude of this effect is not constant, being a function of many variables, including light intensity, relative humidity and leaf water saturation deficit (FRANCO—MAGALHAES 1965, FALK 1966). Finally, the difficulties associated with this technique are not limited to those arising from the excision of leaf tissue. Thus, in view of the unsatisfactory performance of the Piche evaporimeter in a windbreak situation (LOMAS—SCHLESINGER 1971), one cannot help but wonder whether this instrument is really a good choice as a standard evaporatimeter for use in an orchard.

Stomatal density and size were determined with a microscope, this is an acceptable procedure; however, it would have been helpful to have had some indication of the magnification of the microphotographs of these shown in Fig. 3. The collodion technique used for determining stomatal apertures cannot be considered quantitative since, as SLAVIK (1971) has pointed out in a recent review article, stomatal impressions obtained in this way do not necessarily represent the aperture correctly. A further difficulty is that a large part of the range of stomatal aperture going from closed to open lies at the limit of resolution of the optical microscope (0.2—0.3 μ); this is well illustrated by data of COWAN—MILTHORPE (1968a). It seems to me that in both these observations and in their measurements of transpiration intensity, Borka and Borka were really concerned with estimating the diffusive capacity of the leaves, and that they might have done better to replace these observations with such measurements, using a diffusion porometer ("transpiration porometer" of SLAVIK 1971) of which several commercial versions are now available. By so doing, I feel they would have collected more reliable and more quantitative data with probably less expenditure of effort.

The measurements of water saturation deficit (Table 3) have not been expressed in any standard form (BARRS 1968) and so cannot be readily compared with those of other workers.

A considerable effort was made to determine the "forms of water" in the leaves. A perusal of the results section shows that these are considered to comprise "fixed" and "free" or "mobile" water, and furthermore that "free" water is transpired before "fixed" water. This seems to hark back to the concept of the division of cell water into "free" and "bound" fractions, which was rejected as long ago as 1949 by CRAFTS—CURRIER—STOCKING. At the very least I consider such terms as "free" or "fixed" water as archaic and misleading, since in reality there is not, and cannot be, any fraction of water which is "free" in a cell. In a steady state situation, all water in the leaf must be at the same water potential (NOY-MEIR—GINZBURG 1967, WEATHERLEY 1970). As SLATYER (1967) pointed out, it follows that any particular bound water determination reflects nothing more than a point on the water potential *v* water content curve. Clearly, no conclusion as to the relative amounts of "free" and "bound" water can be made, the amount of "free" water measured will depend on the initial water potential of the tissue and on the water potential to which the tissue was exposed during the determination. Probably, what this unfortunate terminology means to convey, is the magnitude of the ratio between the osmotic and the matric volumes (WEATHERLEY 1970). The present results (Table 8), based on inadequate techniques, as explained above, lead to the conclusion that the proportion of "fixed" water greatly exceeds that of "free" water. The rather meagre data available from other researchers using techniques more suitable to the problem, suggest that the reverse is the case (BOYER 1967, WARREN WILSON 1967, WEATHERLEY 1970, WIEBE 1966) in all, except possibly 'hard' leaves such as those of the eucalypt (KREEB 1965). In the present work a determination of the water potential *v* water content curve for each of the three peach varieties might well have been more informative and a better index of their water regimes, than the determination of "free" and "fixed" water. While on the subject of water potential it should be noted that the paper refers to measurements of water potential in Table 3 and states that water saturation deficit is inversely correlated with water potential. This may well be so, however, no actual leaf water potential values are in fact presented.

In considering the techniques used I have inevitably included discussion of some underlying concepts which seem doubtful as to their validity. Another such concept is contained in the statement 'The smaller the stomata, the higher the possibility of vaporization.' According to COWAN—MILTHORPE (1968b) this is a fallacious view, probably arising from failure to recognize the importance of the boundary layer resistance immediately above the stomata, and the subsequent invocation of a vague and equivocal "interference" between stomatal pores. Indeed, Borka and Borka's own data are inconsistant with this concept, since they found that the peach variety with the highest transpiration rate (Julia) had the lowest stomatal density and the largest pores (Tables 2, 6 and 7).

The authors discuss briefly the causes of diurnal plant water deficits and point out the two possibilities that these arise as a result of resistance to liquid phase flow in either the plant (Rp) or in the soil (Rs). They rightly draw attention to Weatherley's view that Rs is primarily responsible (see also MACKLON—WEATHERLEY 1965). However, this is currently a controversial area and they should perhaps also have mentioned the view of NEWMAN (1969a, b) that Rs only becomes dominant when soil water content is at or near the permanent wilting point.

Another interesting point mentioned briefly is their observation that stomatal closure occurred earlier in the day in older than in younger leaves, probably as a result of the more effective competition for water by the younger leaves. In fact this is not the first time such a phenomenon has been reported, and it might have been of interest to cite other reports of differences in water status between older and younger leaves such as have been found by Begg *et al.* (1964) for bulrush millet, by Lemon (1963) for maize, and by Čatskú (1962) for fodder cabbage. Borka and Borka's results are of especial interest since they do not support Cowan—Milphope's suggestion (1968b) that the effect may be due to less responsive stomata in older leaves.

In conclusion, I would say that the topic investigated by Borka and Borka is certainly worthy of study; more data on plant behaviour in the field is urgently needed (Monteith—Biscoe 1971), especially over the life of a crop, as was attempted here. One can only regret that the present study was not undertaken with more contemporary techniques and a more contemporary outlook. It was also disappointing to find a complete lack of any statistical analysis of the data, making an objective comparison of results from the three peach varieties studied very difficult.

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CAN THE COLLODION METHOD BE RELIABLY USED IN DETERMINING THE DIAMETERS OF STOMATA?

Distinction should be made between water circulation and water economy in plants. Water circulation is characterized by motion and forms, a chain of movements consisting of three phases: uptake, transport and emission. The continuity of water circulation is ensured first of all by the water supply. When the water supply is not satisfactory the economy of the existing water content becomes imperative. It is characterized primarily by an increased ability of the cells to retain water, connected with an increase in the water absorbing power; or, to say it with a greater physico-chemical accurateness though less descriptively: with the value of the water potential becoming more negative. Water economy includes the translocation of internal water contents from older to younger leaves, that is the reutilization of water. It is a pity that Gy. Borka's paper does not give a clear picture of these facts.

A very important water circulation index is the extent to which the stomata are open, and though not directly proportional to transpiration, it indicates the turgescence of the guard cells. However, the collodion method mentioned in the paper may mislead the researcher, because the collodion which penetrates into the open stoma hardens wedgewise, therefore the microscopic picture of the diameter of the stoma, as seen on the collodion membrane, depends on the angle of setting. The infiltration method might have been more suitable for studying the aperture of the stomata.

The proline content of leaves which indicates the occurrence of a water shortage is a very characteristic index. A quick method of pointing out proline was elaborated in Hungary by Gábor Pálfi who used isatin impregnated paper for this purpose. Thus it is worth while taking the concentration of free amino acids — especially of proline — in consideration when studying the water economy of plants.

In their paper on water circulation in peach-trees the authors mentioned that they had studied the indices of water economy in leaves of the same age. Among the indices of water economy the paper naturally also mentioned the saturation deficiency. It would be very fruitful to study the gradient developing between differently inserted leaves of the same shoot under the influence of an incipient water deficiency. This is usually manifest in that water is withdrawn from the older leaves with the result that the saturation deficiency increases parallel with the age of the leaves. Occasional irregularities in the gradient originate from the fact that certain leaves may communicate with leaves belonging to the same orthostychon rather than with those closest to them.

Thus, I suggest — in contrast with the authors — that in the course of their study on the water economy of the peach-tree not only leaves of the same age but also younger and older leaves of the same shoot should be compared as to their relative saturation deficiency. In this comparison I should not replace the value of the saturation deficiency by the water potential, as this is probably misleading. We must not forget that the protoplasm and dissolved matter content in the cells of an older leaf are quite different from those in a younger leaf. Consequently, in cells of different age the water potential will be different even if turgor and saturation deficiency were, otherwise, identical. On the other hand, these physiological "parameters" may be different in the case of identical water potentials if the leaves are of different insertion, that is, their age is not the same.

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IS THE INTENSITY OF TRANSPIRATION EXPRESSED AS THE AMOUNT OF TRANSPIRED WATER OVER THAT OF EVAPORATION FROM AN OPEN WATER SURFACE?

This article effectively shows that differences in variety have a marked effect on transpiration and consequently on water regime. In order to achieve this, the authors were careful to measure transpiration on leaves of the same age, since it had previously been shown by different authors that there is a close correlation between transpiration and age. They also studied trees of the same age, growing in identical soil conditions. This contributes to validate their results. Thus they were able to show that transpiration is most intensive in the variety Julia and that in the daily rhythm of transpiration, the variety Julia is the first to obtain a maximum.

However, to get the most of scientific benefit from their work, several other precautions should have been followed. 1° In the graphs, tables and text, all three varieties should have been compared at all times. 2° In the "Results" section, only results from the present investigation should have been included. Results from other workers and possible explanations or interpretations of the observed phenomena should have been consigned under a new heading: "Discussion". This is because the constant inclusion of discussion or interpretations within the "Results" section dilutes the text and prevents the reader from grasping the exact contribution to science by the authors.

Also, in this section, every affirmation should be substantiated by figures. For example, in the following phrase "the total water content determined in the leaves of the variety Győztes at the beginning of September, and the ratio of water forms within showed — as opposed to the variety Elberta — a considerable slowing down of the metabolism (Table 8)". Since the intensity of metabolism has not been evaluated conventionally here, this affirmation should be excluded from this section. Correlations between degree of hydration and metabolism are known, but this should be elaborated in the discussion section.

To the casual reader, some other details should be included. For example, since all readers have not read the previous papers of Borka and of Parcevaux, the basis for expressing the intensity of transpiration as a percentage (Figs 1 to 4) should be stated. One may only suspect that it is the expression of amount of transpired water over that of evaporation from an open water surface.

In order to understand the views of Tew—Taylor—Aschroft (1963), one must realize that transpiration cannot be said to be a function of relative humidity, unless all experiments are carried out at the same temperature. In fact, transpiration is dependant upon water vapour pressure, as expressed in mm of Hg and not upon either relative humidity of temperature alone. For a good text on the subject, read "An introduction to Plant Physiology" by O. F. Curtis and D. G. Clark, McGraw, Hill Book Company, New York, 1950. First edition, p. 191—199.

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WHAT MORE SHOULD WE KNOW ABOUT THE WATER RELATIONS OF PLANTS?

Drs. Gy. and K. Borka have made a useful contribution to an important aspect of fruit culture in their article:

"Daily and annual rhythm of water regime indices in peach varieties of various ripening times".

Despite the practical importance of adequate water for the production of sound, marketable fruit, the subject of indices sensitive to water stress in fruit trees has received all too little attention.

French (1961) investigated the irrigation of fruit trees in Australia for many years and stated, "Current irrigation practice has been very largely evolved from the concept that soil moisture is equally available to the plant between field capacity and wilting range. However, . . . this involves suboptimal (water) levels at which yield and quality may be adversely affected". He considered the effect and nature of these suboptimal levels of soil moisture require urgent study if irrigation practice is to be placed on a rational basis. This, of course, is at variance with Veihmeyer—Hendrickson's (1950) conclusions that the growth of pears, prunes, peaches and apples in California is not affected by changes in soil moisture, until it falls to the permanent wilting percentage.

The findings of Weihmeyer and Hendrickson on deep, well structured Californian soils should only be extrapolated with caution to other field conditions. French (personal communication) has since found after several seasons of precise measurement, that the sizing of peaches is adversely affected by water stress when soil moisture levels are between field capacity and the permanent wilting percentage. He also found water stress on peach trees below the permanent wilting percentage caused fruit shrinkage and a lasting setback to fruit development. This setback was proportional to the length and severity of the period of water stress.

Whilst Drs. Borka do not report on fruit development or tree growth, they do indicate that this will be a later objective in their studies. They found in their present study that water stress could occur in trees even when soils are adequately supplied with water. Their findings are thus similar to those cited for French with respect to fruit development. They observed that the transpiration rate declined towards noon, because of high insolation; and this resulted in an unfavourable water balance in the tree, even when the soil was at field capacity. A rise in leaf temperature accompanied the increased moisture stress at noon. This was indicated by the closure of leaf stomata during the stress. Findings of this character are important and help to explain differences in interpretation of the effectiveness for plant growth of soil moisture within the available range.

In reporting the results of this paper, the authors assumed that the synthetic processes essential to normal development could only proceed if tissue moisture levels were "satisfactory" for high yields. Some expansion of this concept in a quantitative manner would have proved useful to the presentation, even though it is to be a later phase of the investigations. The single statement that the yields of irrigated peaches were 20—40 per cent higher than unirrigated, does not suggest a degree of sensitivity in growth response to tissue moisture stress, such as is later claimed in the paper.

However, GATES (1968) comes to similar conclusions after a resume of a large mass of precise physiological data relating to many species. He concluded that tissue moisture levels must be high if active synthesis is to proceed. OWEN (1952) established precise levels of water potential for the germination of wheat, by placing grains in small chambers under close control. The grains were allowed to come into equilibrium with a range of salt solutions by vapour diffusion alone. Owen found that the rate of growth was directly related to water potential.

GATES (1964) postulated that water may have a biochemical role in relation to macromoleculas in the physiological processes of the plant. Much the same is inferred by Drs. Borka when they claim that changes related to the hydration of colloids took place in the protoplast resulting "in a slowing down of the active physiological processes and an increase in the decomposing enzymes". The daily change in "free" and "mobile" water that they report is obviously relevant to these claims.

The usefulness of these results would be enhanced by the enumeration of statistical significances. On the other hand, it is important to know how water stress exerts its effects upon metabolism. To suggest that there was an increase in the enzymes of decomposition is perhaps descriptive of the effects of moisture stress, but it does not reveal any of the mechanisms involved.

It is of little value to say as did WOOD (1939):

"Water does not exert an effect as such, that is, as a molecular species . . . decrease in water content will only produce effects on metabolism by increasing the concentration of other substances."

Some modification of such a view is now necessary, in the light of modern knowledge of the relation of water to the functioning of the plant protoplast.

Evidence is now coming forward that water may have an effect *per se* for biologically reactive molecules. GATES (1968) suggested that in considering such an effect it is necessary to not only account for the cessation of growth during water stress, but also the ready resumption of development and synthesis that ensues upon restoration of turgor. He suggested (1964), after considering some aspects of the relation between macromolecules and their hydration shells, that the role of water in the protoplast might be that of linking macromolecules into a co-ordinated dynamic whole, perhaps by hydrogen bonding. During water stress, the protoplast might undergo a change of state such that bonding positions are partially satisfied within the macromolecules themselves, with consequent suspension of metabolic activity. Upon rewatering, the change in water activity might be expected to favour the restoration of normal bonding positions. CRAFTS (1968) suggets that such a concept warrants further consideration, and further enlarges on Gates' suggestion that water may have a functional relation with such biologically important macromolecules as DNA and protein.

The knowledge of how the relationship between plant water and the protoplast may occur is extremely limited for obvious reasons. The reversible hydration and dehydration of proteins may alter their architecture and so affect the specific functions of enzymes which, in turn, determine the rates of most metabolic reactions. CRAFTS (1968) quoting MAHMOUD (1965) states that dessication resistance is a property of the plant protoplast and not a mechanical-structural property. This being so, the investigation of the plant protoplast as an organized entity is a desirable, even though extremely difficult, matter. It will not be achieved by descriptive reference to changes related to the hydration of colloids. GATES (1968) suggested that clarification of the relationship between the living plant protoplast and its water content might best be achieved by creating sets of systems of changing metabolic states that are associated with plant ontogeny, and then manipulating these to advantage. With modern techniques of chemical and statistical analysis the matrix of change that results from gradations of treatment might thus be assessed and interpreted, and so the parameters might be determined for the role of tissue moisture in effects of water stress on plant growth.

The paper discussed here is a step in this direction, and provides valuable information regarding changes in water stress in trees in the field, where conditions of experimentation are difficult. The use of modern methods of statistical analysis and especially multivariate analysis would enhance the value of future studies, for these would allow parameters for the role of tissue moisture in development to be assessed with even greater clarity.

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WHAT IS THE RELATION BETWEEN TRANSPIRATION INTENSITY AND WATER UPTAKE?

I have read the manuscript of the paper carefully and with interest. I should like to comment this paper as follows:

Is the paper a survey or an experimental account? If the paper is experimental, the object and methods should be precised accordingly. I recommend the term "water relations" instead of "water regime". Instead of saying that the transpiration intensity was measured according to the method of BORKA (1967) and PARCEVAUX (1964), the correct citation would be according to PARCEVAUX (1964) and BORKA (1967). The methods used for measuring the density and dimensions of the stomata should be described precisely. The particular determined water forms in the leaves should be defined. Tables 1 and 2 should be omitted. The numerous values of the Tables should be given in the text. From how many values is the average calculated in Fig. 1. In Table 3 is 8 m^2 leaf area 160 mg? In the text the absence of differences is stated between the varieties Julia and Elberta regarding the decrease in transpiration resulting from an overcast sky. In the Tables the differences are introduced. The age and position of the leaves where the density and length of stomata were examined should be precised. In the text stoma density is given per cm^2, whereas in the Table it is per m^2. It is unusual to compare the transpiration on the basis of the evaporation from the free water level. The relation between transpiration intensity and water uptake is not clearly formulated. The last section "The water uptake by the plant . . ." is very general and I recommend omitting it.

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UTILIZATION AND FUTURE LINE OF STUDY OF WATER CIRCULATON INDICES IN PEACH VARIETIES?

Gy. Borka—Gy. Borka's paper on "Daily and annual rhythm of water circulation indices in peach varieties with different ripening times" is made more valuable by the fact that the author's test plant — the peach tree — and thus his statements too are of practical importance.

The practical application of research results is promised by the paper itself: "Our further aim will be to study the course of shoot growth and fruit development in these varieties as reflected by the most important indices of water economy . . ."

Accordingly, I should like to make some comments on the subject:

1. The soil and precipitation conditions of dry farming in the traditional peach growing regions of Hungary compelled the growers to practise intensive pruning of peach trees in order to ensure uniform yields of sufficient amount and quality. This tradition of short pruning is maintained — though often in a modified form — in the new peach growing regions too, where the trees display a much more intensive growth vigour which might give an opportunity to introduce — especially under irrigated conditions — the more up-to-date American method of long pruning. Long pruning has gained ground even in France, the country of classical peach pruning, and in the main peach growing regions (e.g. in the Rhone valley) this is now the dominant method of pruning. The essence of the American long pruning method is the following: in trees shaped into the required form the bearing shoots are thinned, that is, superfluous shoots are either cut off at the base or pruned to spurs with two or three buds, while the rest of the shoots are left unpruned. In this way the number of pruning wounds will be reduced and extreme regeneration (producing water shoots) caused by pruning too short as well as disturbances in sectorial nutrient transport prevented. It is much more favourable both from the point of view of plant hygiene and concerning the organic matter economy and early bearing of trees. On bearing shoots left at full length the adequate ratio of leaf/fruit is controlled by a supplementary fruit thinning. The annual regeneration of the bearing surface is ensured by the long (unpruned) bearing shoots bending under the fruit load. Namely, on the top of shoot-arches thus developed "replacement shoots" will emerge to which in the spring of the next year shoots will be cut back. However, in the case of late varieties grown in unirrigated orchards the development of "replacement shoots" sometimes represents a problem even under the rainier conditions of France. Namely, fruits of late varieties remaining for a longer time on the bent shoots are rivals in water- and nutrient demand to the "replacement shoot" developing at the top of the shoot-arch. For this very reason, instead of the replacement shoots missing or developing insufficiently in the late varieties the earlier mentioned renewal spurs with 2—3 buds

serve to provide the bearing shoots of the next year. In any case the thorough methods described in the paper would give the possibility of clarifying the water circulation conditions of applying long pruning in Hungary. For example, it could be found out what the soil-, precipitation- or irrigation conditions are under which long pruning is possible and development of "replacement shoots" ensured — depending on the intensity and course of transpiration in varieties with different ripening times.

2. The paper finds a correlation between fruit development and transpiration intensity. In this context further investigations into the question of fruit quality also become important. Namely, the paper points to a decrease in the photosynthesis as originating from a reduced transpiration intensity caused by water deficiency. On this occasion the idea of a remote connection with an earlier experiment presents itself. Manaresi (cit. BREVIGLIERI, N. 1950: Peschicoltura. 590) studied in an ingenious experiment the correlation between the length of the nutrient sucking shoot next to the peach fruit and the development of fruit components. In the case of one-third of the fruits examined the nutrient sucking shoot was removed, in the case of one-third it was cut back to half-length and in the rest of the cases left at full length. The result was the following: in fruits completely deprived of nutrient sucking shoots the water content, while in those with nutrient sucking shoots left at full length the dry matter content was too high; the proper balance of components was found in fruits where the nutrient sucking shoots were cut back to half length. The obvious reason for all this was that in the above treatments the influence on the fruit of the nutrient sucking shoot was different, that is, the nutrient sucking shoots provided assimilates for the fruits with photosynthetizing surfaces of different size. Now, extrapolating the conclusions drawn from Manaresi's observations, I suggest that by means of the methods described in the paper, correlations should be found between the intensity and course of transpiration in peach varieties of different ripening time and the leaf/fruit ratio ensuring the proper balance of fruit components, as well as the best shape and colour in the different varieties.

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DOES THE HIGHEST RESISTANCE IN WATER MOVEMENT OCCUR WHEN THE WATER MOLECULES ENTER THE ATMOSPHERE THROUGH THE STOMATA?

In this review, the writer will consider the present paper by Borka and Borka in three sections; firstly as a scientific paper, secondly with respect to the experimental methods and lastly, the results and discussion will be treated.

As a paper, it is very disappointing, partly because only half of the story of these investigations is given. To illustrate this point, in Figure 4 we are presented with some very interesting data on varietal differences in transpiration rate during the growing season. However, these results would have been much more revealing if details of the resulting development and yield of fruit had also been published. We are promised this information at a later date in the Introduction to the paper but the reader would certainly have benefitted from the simultaneous publication of these two sets of data.

In addition, the fragmentary nature of the results presented in Tables 1 to 8 does not do justice to the work of the authors. An outstanding case of this is Table 4 where two leaf temperatures are tabulated without any clear indication of date, time of day, air temperature, wind speed or leaf age. Presumably, as a result of two seasons' experimentation, there must exist a large body of observations from which Borka and Borka have selected a very limited number of values. As a result, the reader is denied an overall view of this research and, more important, the opportunity to compare the work with his own experience. The reviewer is not suggesting that the paper should become merely a catalogue of experimental results; however, a judicial use of appendices containing a more representative sample of data would aid the communication of ideas. For instance, many workers would have appreciated the tabulation of a comparison of water regime indices and leaf temperatures between the time when soil water supply was adequate (70% capacity) and the latter half of July 'when turgor was not re-established in the leaves even during the night'.

However, the most crucial criticism of this work as a scientific paper, which should have excluded it from any scientific journal, is the fact that no standard errors or confidence limits are assigned to any tabulated value or graph. Consequently, assessment of the significance of inter-varietal differences, for instance, is not possible.

In the short time allotted for the preparation of this article, the author has not been able to become thoroughly acquainted with all the methods of measurement of water indices employed by Borka and Borka. This is particularly the case for the measurement of transpiration rate using a thermister transpirometer as described by GY. BORKA in his thesis (1967). This thesis has not become available to the reviewer nor has any other account come to hand of this type of instrument. One must assume that this method involves the estimation of the water vapour content of air issuing from a transpiring leaf by a development of the traditional wet and dry bulb thermometer method. It would probably be most valuable to crop physiologists and ecologists if Borka were to publish details of his transpirometer.

The study of 'forms of water' in a plant leaf is difficult to discuss since such 'forms' defy definition and anatomical location. The present authors follow the ideas of GUSEV (1965) who measured water removal from tissues by solutions of increasing osmotic pressure. These measurements have yielded interesting results as shown in Table 5; however, as SLATYER (1966) has shown such data may be subject to the magnitude of the appropriate reflection coefficient of the solute used to increase osmotic pressure. In addition, some allowance must be made for solute movements to and from the leaf tissue.

When Borka and Borka measure the 'absorption value' of leaves, this parameter is identical to suction tension, diffusion pressure deficit (DPD) or, most important, water potential (KOZLOWSKI 1968; SLATYER 1967). Their method, based on ÖNÁL (1964), involves the equilibration of leaf tissue with the vapour of solutions of different osmotic pressures. If performed under optimum conditions of temperature control etc., the method is adequate (SLATYER 1958, 1967). However, much better reliability may be obtained by using some form of psychrometer (SLATYER 1967) although this may be expensive.

Recently, a very comprehensive review of methods of measurement of leaf saturation deficit has been published (BARRS 1968) and the reader should consider the present method in the light of that paper. However, two outstanding points must be raised with respect to the results of Table 3. Firstly, the expression of deficits in terms of leaf surface area is to be discouraged since leaf area may alter with water stress; secondly, equilibration of the tissue for only 60 minutes may not be long enough. The reviewer is here handicapped since the original quoted article by BORKA (1969) was available only in Hungarian. (One assumes that Table 3 is expressed as 'mg weight of 8 cm^2 leaf' and not '8 cm^2' as stated. See also Table 6.)

The writer is still in doubt as to the measurement of 'the course of water loss in the plant' since no such account is given in de PARCEVAUX's (1964) largely theoretical paper. The stomatal studies and leaf temperature measurements appear to have been performed in conventional ways.

The results obtained using these methods agree well in several ways with the work of other authors on crop plants in the field. In particular, Borka and Borka find in their material the widespread phenomenon of 'midday closure' of stomata (MEIDNER—MANSFIELD 1968); similarly, they find that transpiration rate is a function of the insertion and age of leaves and that leaf water potential is inversely correlated with leaf water saturation deficit. However, this last correlation is drawn from unpublished data, as is the statement — 'the nearer soil moisture approached the total water capacity, the higher was the number of open stomata found on the leaves'. The authors must realize that the critical reader must be provided with all data relevant to conclusions.

The views of the authors on the site of highest resistance to water vapour movement from the leaf require some clarification. They consider that 'the highest resistance in water movement occurs when the water molecules enter the atmosphere through the stomata. The smaller the stomata the higher the possibility of vaporization'. This is a misrepresentation of the facts. It has been shown clearly (BANGE 1953, MEIDNER—MANSFIELD 1968) that, in still air, the highest resistance to water vapour movement from the transpiring leaf is the diffusive resistance of the air external to the leaf. Under such conditions, stomatal aperture has very little controlling effect upon transpiration rate. On the other hand under normal field conditions there is considerable movement of air causing a reduction in the resistance of the air external to the leaf. In consequence, the movement of water vapour through the stomatal tube becomes rate limiting and the size of the stomatal aperture controls transpiration rate. Thus, in contrast to the statement of Borka and Borka, the smaller the stomatal aperture the lower will be the transpiration rate under normal field conditions.

A further point worth mentioning at this point is that the authors attribute the midday decrease in transpiration rate to 'reduced turgor'. Decrease in turgor under these conditions causes the closure of stomata and, therefore, reduction in transpiration rate; clearly, this reduction is an indirect and not a direct consequence of the decline in leaf turgor.

To the reviewer, the most interesting and valuable data in this paper are contained in Figure 4 which shows inter-varietal differences in transpiration rate during the growing season. For each variety, the three phases of water utilization may be recognized, always assuming that the features of the curves are significant. It appears that the variety Győztes has completed the phase of stone hardening (and maximum transpirational water requirement) before the onset of drought in the second half of July. In contrast, the variety Julia required most water precisely when least was available. At first sight, it would seem that, as regards water utilization at least, the variety Győztes is better adapted to Hungarian conditions. Further consideration of the varieties is not possible without details of the development and yield of fruit. In addition to yield, fruit 'quality' may also be a factor worth taking into account since, for example, KRAMER (1963) quotes a case of the 'quality' of apricots and pears being improved by 'water stress late in the growing season'.

In conclusion, it seems that Borka and Borka are working with material which can yield not only some very interesting results but also clear indications of how water resources should be managed to secure optimum yields. In general, they have used reasonable experimental methods and they have described their experiments in good English. However, the authors are urged to do more justice to themselves, their experimental methods and their material by improving the standard of presentation of results as outlined in this review. In addition, improvement of the method for the determination of water saturation deficit is necessary, and the authors might consider studying the movement of water to and from fruit, since some workers consider this to be of importance (see SLATYER 1967, KOZLOWSKI 1968). The reviewer looks forward with interest to the publication of the results of the irrigation experiments.

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HOW IS TRANSPIRATION CONTROLLED BY TURGOR?

The paper by Borka and Borka is quite interesting, but there are a number of faults. No standard errors are given for the data in any of the tables or figures. It is therefore impossible to judge which of the results are statistically significant and hence it is impossible to come to a valid conclusion.

The analysis given on transpiration does not seem to be sufficiently rigorous. Reference is made to transpiration being controlled by turgor. Turgor can only affect transpiration indirectly through altering stomatal aperture and it is in these terms that the analysis should be made.

Table 3 — expressing saturation deficient in terms of weight per unit leaf area seems to me curious and perhaps not entirely reliable.

Table 4 — single values of temperature without indications of errors or changes in time seem of little value.

Table 5 — I am not sure how these percentages of free and fixed water were found, but is seems to me that such high percentages of fixed water are rather strange.

Table 6 — stomatal frequency is given as numbers per m squared. Surely this should read per cm squared.

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WHAT IS THE RELATION BETWEEN SOIL MOISTURE AND THE OPENING OF STOMATA?

The paper by Borka and Borka deals with an interesting ecophysiological problem deserving publication. Nontheless there are some points, to be commented upon.

In the opening part of the paper a summary of results is presented. This appears to include some statements not fully supported by the data presented in the Results.

1. Fig. 1 does not seem to justify the statement that "towards noon, transpiration decreases in all three varieties due to the absence of turgor".

2. There are practically no data to the conclusion "The water saturation deficit measured at different parts of the day is in inverse correlation with the water potential"; Table 3 referred to in this connection only contains data on water uptake by leaves (or leaf disks?).

3. The meaning of the subsequent sentence "At noon, with total insolation, the temperature of the leaves suffering from a water saturation deficit is 4—5 °C higher than at the stage of total turgor" is obscure. The same is true about the relevant data of Table 4. The confusion could be avoided if data for all four combinations of factors considered (sun and shade, full turgor and water deficiency) were presented.

4. No data are given to demonstrate the relation between soil moisture and the opening of stomata.

The remarks 1 to 4 pertain above all to the selection and interpretation of data.

The presentation of data also deserves some comment. The informative value of the paper could be considerably increased if the Tables were supplemented by additional data on numbers of measurements, sizes of samples, etc., as well as by some statistics such, as standard deviations, fiducial limits, and/or *t*-tests in case of differences. It would be also desirable to provide Tables and Figures with dates wherever possible. The correlation coefficient for the relation between number and size of the stomata could be presented in the Results. Generally, the same holds true for the Figures as for the Tables. Here, eventually, the individual data could be plotted to make the reader acquainted with the extent of their unavoidable scattering. With all this additional evidence not available it is difficult to discuss some of the results obtained.

Personally, I would prefer subdividing the section Results in proper Results and a Discussion, so that a clear distinction between experimental findings and generalized explanations of these would be easier than with the present arrangement of the paper.

Unfortunately, I am not familiar with the methods by BORKA (1967) and PARCEVEUX (1964) mentioned in Material and method. Therefore I can only suggest that the term "relative transpiration" should be preferred to "transpiration intensity" if it is not the absolute transpiration that was measured.

The statement: "The young leaves were less xerphilous than the older ones" deserves a more thorough discussion, as generally the opposite is accepted (e.g., the law of Zalenski).

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IN A LEAF CONTAINING 70—80% OF WATER HOW MUCH OF IT IS IN A COMBINED STATE?

I have read with interest the treatise entitled: "Daily and annual rhythm of water regime indices in peach varieties of various ripening time" by Gy. Borka and K. Borka, published in Acta Agronomica.

Apart from light, water is undoubtedly the most important factor in vegetable production and, therefore, research work concerning the consumption and utilization of water by cultivated plants deserves general attention.

Under the conditions of Hungarian pomiculture, the choice of the peach as the object of research is certainly reasonable. Researches in natural conditions are most indicated and require the use of rather simple and quick methods. Their choice depends on the author's interests and on the available apparatus.

The decided division of the forms of water in the plant tissues into "free" and "combined" water may give rise to some doubts, the more so, as the amounts of "combined" water are enormous. I should like to draw the authors' and the readers' attention to the book by G. Hübner, K. Jung and E. Winkler, published by "Akademie-Verlag GmbH" in Berlin in 1970, in the series "Wissenschaftliche Taschenbücher". The book is worked out on the basis of extensive literature (371 items). It concludes from various precise physical measurements that the amount of water closely combined with 1 g of nucleic acids is 0.1—0.2 g, and of that combined with proteins — 0.2—0.5 g. In the plant cell the water content is very high. Though a part of it may be combined as a result of imbibition, the sucking force of the swelling colloids rapidly diminishes as the water is absorbed. In a cell containing 80—90% of water the colloids are nearly completely saturated with water.

Although in the concept of "water potential" its component part, the so-called "matric potential" resulting from the sucking force of the colloidal substances in the cell, is taken into consideration — this force is so insignificant that it is considered rather on account of theoretical accuracy than for any practical purpose.

The sucking force of the cell is due chiefly to the diffusion pressure deficit. It is difficult to imagine that in a leaf containing 70—80% of water (Table 8) 90% of it should be in the combined state. Is the water in the vacuole, which contains a half of the cell water, also combined and if so — with what?

This is an essential question arising when one considers the concept of combined or fixed water. The problem seems to be controversial, and therefore a too unhesitating and arbitrary division of cell water into free and combined (fixed) water, a division based on one simple method, may be open to doubt.

When reading the treatise, the following small questions occured to me, calling for the Authors' explanation:

When discussing the results, the authors write: "Transpiration is related to the total water content of plants; waters fixed the least are the first to evaporate, and colloidally or osmotically fixed forms of water are moved only in the absence of free water". This conclusion calls for experimental justification.

The sentence: "Later on, around noon, when the turgor decreased, the water deficiency in the leaves increased" should be formulated differently, for it is not the decrease of turgor that provokes the deficiency of water, but quite conversely: the turgor decreases due to water deficiency.

Further, the sentence: "The smaller the stomata, the higher the possibility of evaporation" also gives rise to some reservations. An attentive reader will understand that this is only a mental abbreviation. Greater possibilities of transpiration result from the greater number of stomata, but not from their small dimensions. This would contradict the basic laws of diffusion and, without additional explanation, may lead a less critical reader into error.

I have also certain objections as regards the final remarks; one should not speak about enzymatic activity which has not been examined, nor about the passing of the plant into the stage of dormancy owing to the exhaustion of energy reserves. The authors have not been engaged in researches of this kind. Before winter the plant accumulates considerable amounts of organic compounds which constitute a reserve of chemical energy. May be, the authors have in mind the compounds of ATP type, rich in energy, but have not investigated this matter. In Tables 3 and 6 both the mass of the leaf and the number of stomata should have been calculated per sq. cm and not for sq. m. In Table 6 the number of stomata should have been given in round hundreds and their length — in full micrometers. The accuracy with which these dimensions are given does not correspond with that of our research methods and is only the result of mechanical conversion.

Despite certain weak points, the work is valuable; it concerns an important problem and is certainly conscientiously done. The authors attempt to work out accurately the water economy in the leaves of 3 varieties of peaches in Hungarian climatic conditions. Their work shows the substantial differences between the varieties investigated, which can be of importance and have practical application in peach breeding and cultivation.

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HOW ARE ABSORPTION AND TRANSPIRATION LINKED TO RESPIRATORY ENERGY?

I have read the paper by Borka and Borka on "Daily and annual rhythm of water regime indices in peach varieties, etc." I think the paper is weak because the data are not analyzed statistically. Therefore, the conclusions may be questioned. Also the methods are very poorly described. For example, I think it is important to know precisely how stomatal frequency was measured, on how many leaves, etc.

I consider the paper to be much too wordy and inaccurate. I think that absorption and transpiration are not linked to respiratory energy in the way suggested here. The conclusions go beyond the data. I really believe the paper is poorly written and should not be accepted for publication in its present form.

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IS THE NUMBER OF STOMATA PER UNIT LEAF AREA THE SAME ON THE LOWER AS ON THE UPPER LEAVES OF THE SHOOT?

With measuring made at the same stage of development, the lower degree of transpiration in earlier developed peach leaves compared to those developed later — e.g., to the tenth leaf — suggests the idea of a difference in the number and size of stomata per unit leaf area possibly existing between the lower (lower values) and upper leaves of the shoot. This can be easily made certain by comparative histological studies of preparations made of the leaves. The higher number of stomata on the upper leaves of the shoot may to some extent be in connection with the larger dimensions of the shoot axis and the elongated pathways of transportation which involve a more intensive suction force.

There were differences found in transpiration intensity measured at the same time in leaves of different insertion — in the present case in the basal and terminal leaves: at the basal part of the shoot the stomata of the leaves closed earlier. In the upper leaves exposed to a higher insolation the more intensive transpiration is further increased by the thin layer of cuticle which is in connection with their earlier stage of development, not to mention the fact that in the water system kept together by cohesion there is a higher loss of water in the terminal leaves, and from the other parts of the plant, thus from the lower levels, water flows toward the terminal leaves. From the lower leaves water is also abstracted by the upper leaves through the latter's protein colloids and mainly through the energy production of their more intensive respiration. The relatively larger volume of cytoplasm in the cells of the upper leaves compared to those in the lower leaves has also a part in the phenomenon; due to the K ions the water retention and water economy of the cytoplasm is better in the younger leaves than in the older ones which contain more Ca.

The lower degree of transpiration in the variety "Győztes" can be brought into connection with the higher number of stomata per unit leaf area and their smaller diameter; namely in a state of closely spaced stomata the area of covered surfaces between the stomata decreases, the diffusion currents of the stomata meet and clash thus retarding the departure of each other's diffusing particles and — as a result — decreasing the rate of diffusion.

Difference in transpiration between the varieties can be explained not only by the intercellular but also by the epidermal transpiration. On the photos of Fig. 3 the cuticular coverage of the epidermis seems to be varied; this can be clarified by histological examination.

The difference in transpiration between the lower and upper leaves of the same variety, as well as between leaves of different varieties can be in connection not only with the structure of the epidermis but with that of the inner tissues too. While a 10—15 per cent increase in the thickness of leaves is hardly — if at all — visible to the naked eye, this difference in diameter may cause changes in the functioning — e.g. transpiration — of the leaves. Here one may think of the smaller or larger volume of the spongious parenchyma, of palisade parenchyma cells increasing in size without any increase in the number of cell-rows, and of the increasing or decreasing number and space of intercellulars; any change in these conditions may cause differences in the transpiration intensity of the leaves within and between the varieties. No true picture of the extent of transpiration is obtained with only the unit area of the leaf surface taken into consideration and the above conditions (leaf thickness, etc.) left unobserved.

In the period of fruit development a decreased transpiration in the third phase as compared to the intensive transpiration of the second phase is an interesting phenomenon. Simultaneously with the hardening of the stone in the second phase intensive division and organic matter incrustation take place in the fruit; however, in the third phase, at the time of fruit thickening, when the cells considerably elongate and the surface of the fruit becomes much larger — both involving an increased transpiration — almost the same extent of transpiration could be expected as in the second phase. Nevertheless, it must also be taken in consideration that in the first and second phase the green fruit still assimilates and that this may be accompanied by a more intensive transpiration, unlike the yellowing fruit of the third phase. Last but not least, at the final stage of fruit development the hairs that cover the fruit — I think — lose their plasm content completely, and thus their role in decreasing the transpiration increases. Another point here is a probable difference in fruit hairiness existing between the varieties. If this difference does exist, then it has a certain influence on the transpiration intensity of the fruits in the different varieties.

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CAN THE FACT THAT THE TRANSPIRATION RATE SLOWS DOWN WITH THE INCREASING AGE OF THE LEAF BE BROUGHT INTO CONNECTION WITH A SLOWER METABOLISM?

The authors start from the assumption that under the climatic and edaphic conditions given at the place of investigation the productivity of the peach depends on the water relations. Therefore they undertook to determine several indices of water balance of leaves taken from three varieties of peach under field conditions. Characteristic changes in rel. transpiration and water state depending on climatic factors and development were stated. Moreover, differences were found between the varieties regarding water relations. Unfortunately, the authors did not discuss the question, if and by what way their results could be used in order to influence the produce. The interpretation of tables and figures is rendered difficult by the absence of important data, such as absolute transpiration, date, time of day, soil moisture, temperature and other climatic conditions. The number of measurements and statistical values are not given. Some results mentioned in the text are not expressed quantitatively. In Table 3 the data on the water potential and in Table 4 the data on the temperature difference of the water saturated leaves are missing. It is questionable, whether the fact that the transpiration rate slows down with the increasing age of the leaf, could be brought into connection with a slower metabolism, since it is simpler to assume a restriction of transpiration due to an ontogenetically acquired xerophilic nature.

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WHY AREN'T THE METHODICAL DESCRIPTIONS MORE EXACT?

The paper represents an extensive report on experiments dealing with the comparative study of water relations on three varieties of peach trees.

My main critical comments deal with the style of the paper.

The description of the methods is insufficient. The techniques used are quoted by references, but no data on number of leaves, sampling techniques, calculations, reference values etc. may be found.

The chapter Results is a mix-up of description of results (very often superficial) and a too general discussion of these results on the basis of references, many of which are accidental. There are no data on the statistical significance of the results or on absolute values.

Some of the uncertainty and indistinct formulations are probably caused by unskilled translation and not proper English terms (e.g. openess of the stomata instead of aperture, waters fixed the least instead of bound water or free water, tenth storey instead of level, peach instead of peach tree and many more), but most of them are due to inexact formulations, not well defined and properly described results, which can hardly be followed by the reader, who has no fixed points in insufficient methodical descriptions.

All this criticism does not mean that the results may not be interesting, if properly used and discussed. It means only that the paper in the present form may hardly be useful for the reader.

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WHAT ARE THE DIFFERENCES AMONG THE HORTICULTURAL VARIETIES IN THE FUNDAMENTAL PHYSIOLOGICAL PROCESSES?

This paper by Borka and Borka provides evidence for a well-known situation among horticultural varieties, that is that there are differences between varieties in the fundamental physiological processes, exemplified here by transpiration, as well as differences in time of breaking of dormancy, time of blossoming, time of fruit ripening, etc. The data presented on three peach varieties should be useful to growers and nurserymen in peach growing areas. Often the relations of insolation, relative humidity, temperature and depth and water holding capacity of the soil determine which localities in a country are suitable for growing certain peach varieties. It is the working out of these factors, so that satisfactory crops may be obtained, that characterizes the horticulture of important fruit growing districts. This work of Borka and Borka should prove valuable to the horticulture of Hungary.

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CAN THE WATER CONTENT MEASURED IN A STATE OF INCOMPLETE TURGOR EXPRESS THE WATER DEFICIENCY?

All physiological changes correlated with the total water potential are suitable for the indirect determination of water deficiency.

It has been proved by many that water deficiency cannot be characterized when only the water content is known. The water content in the plant organs can easily be determined, it is a great problem, however, that the water contents measured can only be compared with an adequate basis of reference. Reference made to fresh weight, dry weight, dry weight of residue from carbohydrate extraction, total nitrogen or leaf area involves the possibility of these values changing during the investigation and their changes being differently influenced by the numerous external and internal factors.

The water content measured in the state of complete turgor alone can be accepted as the indicator of water deficiency in a given plant. The water deficiency of the examined plants can be characterized either by the value of water saturation deficit (WSD) or by that of relative water content (RWC).

Owing to permanent difficulties in the terminology of water relation, the proper thing to do for the authors is to strictly define the terms they use.

The same stands for the terms: "free" and "bound" water.

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Múzeum krt. 4/a

IS PLANT GROWTH DETERMINED BY INTERNAL WATER BALANCE AND TURGOR IN THE CELLS OR BY EXTERNAL FACTORS?

The water movement in plants and their water balance are influenced partly by external factors, partly by internal characteristics. We should like to add to the statements made by Borka—Borka (1972) that the effects of external and internal factors influencing the water movement in plants cannot be separated from one another. The external factors (meteorological, edaphic, etc.) usually play a definite role. This addition is considered to be especially important because of the following statement made by the authors (Borka—Borka 1972): "The growth of plants is controlled in fact by the internal water balance and the turgor in the cells. These two factors determine mostly the physiological processes and conditions which result in the rate and quality of plant growth and finally in high yields."

In our view plant growth is determined by external factors, which result in the internal water balance and the turgor in the cells, although the plant tries to control both of them. In the case of sufficient water supply and at a low rate of potential evapotranspiration the plant adjusts its water balance optimally, while at a soil moisture level near the wilting point and intensive potential evapotranspiration its water balance will be upset, its turgor is lost until finally complete wilting sets in.

On this basis it is obvious that the first sentence of the above quotation needs supplementation. In our view plant growth is controlled by external factors, simultaneously influencing the internal water balance and the turgor in the cells. According to our formulation the complex of external factors represents the reason, while the water balance of the plant is the result of these factors, in contradiction to the statement in the paper (Borka—Borka 1972), that the water balance is the cause and plant growth is the result. However, in addition to the internal water balance and turgor, the growth is highly influenced by the light and temperature; in fact, sometimes these two factors are the determining ones. It is one of the main characteristics of climatic conditions in Hungary that early autumn and late spring frosts may destroy the plant, in spite of an optimum water balance. It is a well-known fact in Hungary too that in rainy, cold seasons poor in sunshine the crop yields are low and of bad quality, though the water balance and turgor are optimal perhaps throughout the whole growing season. It is precisely at this time, when drought ensures simultaneous high light- and heat energies that outstanding results can be achieved by irrigation.

We think that in the case of peach crops the role of light should be especially emphasized, since one of the purposes of pruning is to supply sufficient light. If we want to emphasize the role of water movement, then the confirmation that with sufficient light, heat and nutrient supply in a given plant variety, the growth of plants is controlled by the internal water balance dependent on the water supply and by the turgor in the cells — has a more general validity. In fact, light- and heat energy, water- and nutrient supply which determine the growth form a dialectic unity, are all equally necessary for ensuring optimum growth, good quality and maximum yield.

It is obvious that studying the effect of any of the major external factors (light, heat, water, nutrients) on plant growth in a series of experiments, all the other factors should be identical. It is only then that the effect the above factors have on growth and yield can be unambiguously determined. Thus, when studying the water balance of fruit trees the following treatments should be set up:

a) The soil moisture (water supply) is changed using a selected variety grown under identical meteorological conditions (light-, heat-, moisture- and CO_2 supply at a given geographical location) and with identical edaphic factors (soil, cultivation, fertilization, etc.).

This experiment answers the question how water influences the growth under given environmental factors (research aimed at determining the necessary amount of irrigation water and the optimum time of irrigation).

b) Under identical meteorological, soil and water supply conditions the nutrient supply of the variety is changed.

Measuring continuously the water use (evapotranspiration) with an evapotranspirometer, lysimeter or measurements on soil water balance, we are able to answer the question of how the different levels of nutrient supply change the water economy (balance) of plants, and through this, their growth (research aimed at determining the effect of irrigation on nutrient utilization, and that of nutrients on water utilization i.e. reduced water movement in the plant, respectively).

c) With identical light-, water- and nutrient supply in a given variety the temperature conditions are changed (glass-house, foil tent, phytotron, mulching).

With consumptive use (evapotranspiration, water use) measured under such experimental conditions, the effect of heat energy on water use and growth can be clarified (research work on acclimatization, breeding, growing under artificial conditions).

d) Heat-, water- and nutrient supply are identical, while light conditions are changed using a given variety (glasshouse and phytotron experiments).

These experiments give an answer to the question of how light influences water movement in the plant and growth if there is a sufficient heat-, water- and nutrient supply (research work of acclimatization and breeding).

It should be noted that while the first question can be answered by field experiments, the last two tasks — with the exception of mulching and soil heating techniques — can only be solved under artificial conditions (phytotron, glasshouse, foil tent, etc.).

If Gy. Borka and K. Borka want to start field experiments to solve further tasks set in the paper (1972) — namely, to study the shoot growth and fruit development of the peach varieties mentioned under irrigated and nonirrigated conditions, as reflected by the major indices of water balance — they have to carry on an experimental series of several years, since — in accordance with what have been said — in a given growing season the shoot growth and fruit development are only partly dependent on the change in the water balance indices. For example, although the amount of irrigation water supplied is the same, in years with different meteorological conditions considerable differences may occur both in the shoot growth and in the quantity and quality of fruit.

The above statement has also been proved by the field experiments carried on for ten years in the agrometeorological observatory at Szarvas. In order to disclose the relations between the water movement of plants and meteorological factors, the following two questions were dealt with in detail:

1. How is the water use of crops influenced by the annual changes of weather?
2. What influence has the plant variety itself on the change in water use?

The essential role of the meteorological factors in influencing the change of water use of crops can be clearly seen when the consumptive use of a plant variety is compared in the different years under identical water- and nutrient supply. For this study maize (MV-1 hybrid)

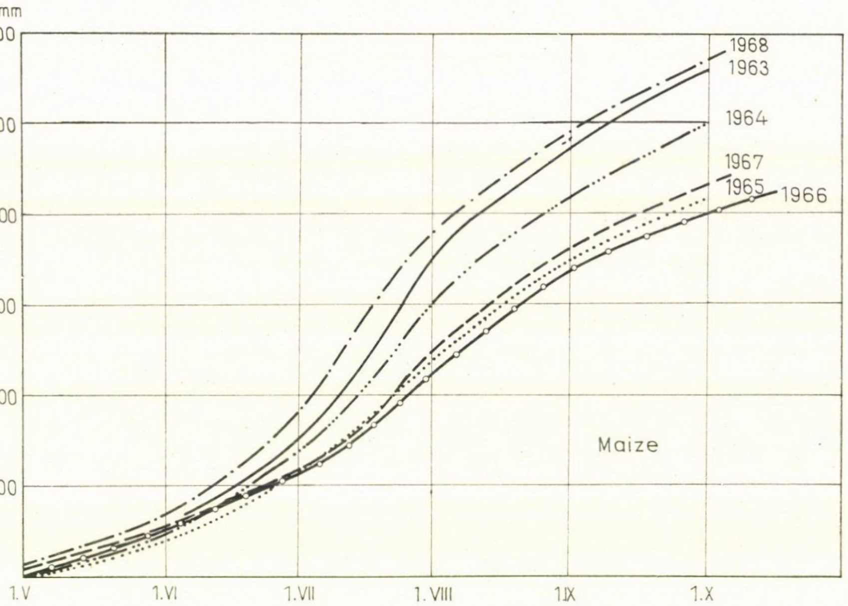

Fig. 1. Summarized evapotranspiration of maize in different years (Szarvas, meadow clay, variety: MV-1)

and potatoes (Kisvárdai rózsa) were chosen. The agricultural technique (cultivation, spacing, nutrient- and water supply, etc.) were identical each year, but the meteorological factors changed from year to year (Fig. 1, 2). The consumptive use of the crops was measured by means of compensation evapotranspirometers (ANTAL 1966).

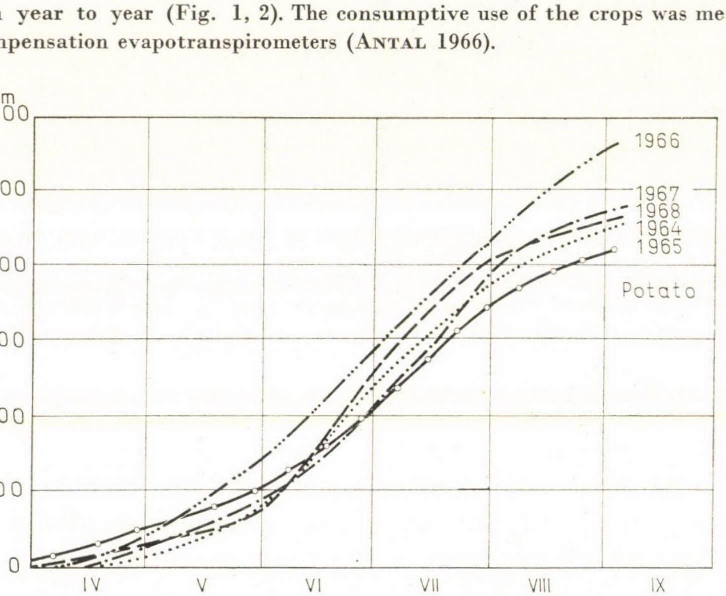

Fig. 2. Summarized evapotranspiration of potatoes in different years (Szarvas, meadow clay, variety: Kisvárdai rózsa)

The summarized curves of Fig. 1 and 2 clearly show that the change of water use in the different years is considerably influenced by the meteorological factors. In the case of both maize and potatoes their water requirements were 30—40 per cent higher in dry vegetation periods than in cool rainy growing seasons. This statement is confirmed even better by the curves in Fig. 3 and 4 which show the changes of daily water use (evapotranspiration) of maize and potatoes in years with different weather conditions.

According to Fig. 4 the daily evapotranspiration of potatoes between planting and emergence is about 1.5—2.0 mm, which agrees with the evaporation of the bare soil. At the beginning of June when potatoes begin to grow rapidly, the daily consumptive use increases quickly too and reaches its maximum at the time of flowering and tuber formation. At that time the average daily evapotranspiration is 5—6 mm; on hot, dry days it may even exceed 8 mm, while in cool weather it decreases to 2—3 mm. After flowering the water requirement decreases sharply, and in the period of ripening it is only 1—3 mm. Similar observations can be made on the change in the consumptive use of maize from year to year and from season to season presented in Fig. 3 with the exception that daily changes in weather do not cause such essential decreases in the daily evapotranspiration.

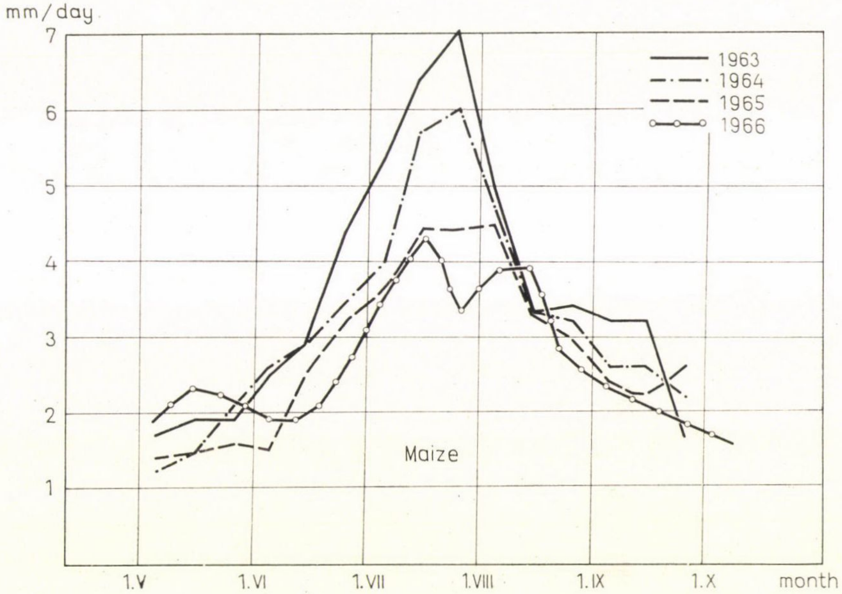

Fig. 3. Changes in the daily water use of maize during the growing season (Szarvas, meadow clay, variety: MV-1)

The data in Table 1 also prove the decisive effects of weather conditions. The data in the Table present the daily water use values of potatoes and maize growing in evapotranspirometers in the period of flowering and tasselling, respectively, that is at the same phase of growth, on days close to each other but with different temperatures.

According to the data of Table 1, in 1964 and 1965 under the influence of a 7 and 10 degree-fall in the temperature, the water consumptive use decreased by 50—80 per cent in

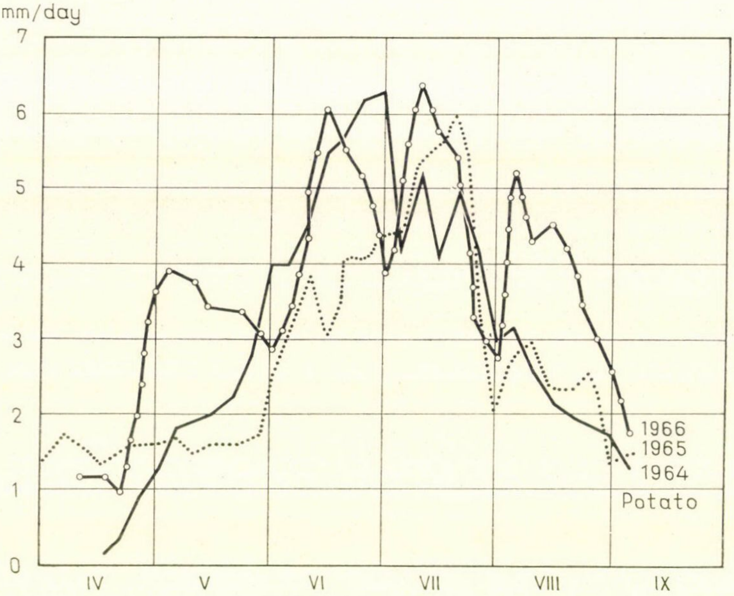

Fig. 4. Changes in the daily water use of potatoes during the growing season in different years (Szarvas, meadow clay, variety: Kisvárdai rózsa)

Table 1

Changes in the daily water use of the potatoes (Kisvárdai rózsa) and maize (MV-1) on days with sharp variations in the air temperature (Szarvas, meadow clay soil)

Time	Water use of potatoes mm	Daily average air temperature °C	Time	Water use of maize mm	Daily average air temperature °C
1964 June 28	7.1	24.4	1963 July 26	10.0	25.9
29	7.7	23.2	29	2.3	17.5
30	4.3	14.5	30	3.5	17.8
1965 June 24	6.2	23.4	1964 July 22	9.4	27.1
25	6.4	24.4	24	3.1	20.8
28	2.6	16.5	25	3.6	19.7
1966 June 30	2.8	14.4	1965 July 26	7.1	22.6
July 1	2.4	16.7	27	2.4	16.9
5	7.1	24.6	28	3.7	18.0
1967 June 8	7.0	22.0	1966 July 20	7.3	24.0
9	1.3	17.3	23	1.7	17.0
10	1.4	16.5	1967 Aug. 7	2.2	18.8
11	0.6	11.4	10	8.2	25,3
1968 June 9	7.4	22.9	1968 July 15	11.0	24.7
10	2.5	13.1	18	2.5	17.1
11	2.0	12.4	19	2.7	16.1

24 hours. The opposite was found in 1966, when an 8° rise in the temperature increased the daily water requirement by 70 per cent, though there was no change in the water supply.

Between 1963 and 1968, under the influence of a 6—10° decrease of temperature, the daily evapotranspiration of maize fell back to one fifth, compared with the days of maximum water use. The same rise in temperature caused a three- or four-fold increase in the water requirement of the crops (see the change from 7th to 10th August).

On the basis of the above mentioned we may state unambiguously that the annual variability of weather causes considerable fluctuations in the water use of crops during the growing season, even if they are optimally supplied with water.

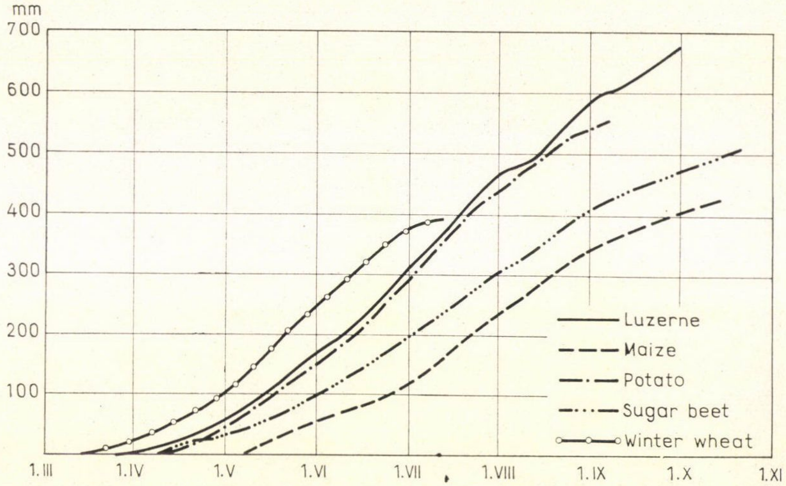

Fig. 5. Summarized evapotranspiration of different crops in 1966 (Szarvas, meadow clay, varieties: alfalfa Szarvasi kékvirágú; maize MV-1; potatoes Kisvárdai rózsa; sugar-beet beta-poly 2; winter-wheat Besostaya)

Gy. Borka and K. Borka point out that variety and growth phases are also decisive factors in the water balance of the peach tree; to prove their statement they present tables and diagrams. The effect, variety has on water requirement and water use can only be determined unambiguously when we compare the water balances of different varieties growing under identical meteorological conditions.

Our own results also support the above statement — however, the results were obtained with field crops and not with peach trees. Our experiments were carried out at Szarvas with different varieties of potatoes (Kisvárdai rózsa and Gülbaba), and maize (Mv-1 hybrid, Mv-620 hybrid, as well as with the Yugoslavian maize variety ZPSK-6 hybrid). Considering that the differences in the water use of the varieties are less remarkable than in the water use of the species, we are presenting some results related to the latter.

Fig. 5 shows the summarized curves of the evapotranspiration of alfalfa (Szarvasi kékvirágú), potatoes (Kisvárdai rózsa), winter wheat (Besostaya), sugar beet (Beta poly 2), and maize (Mz-1 hybrid) with optimum water supply in 1966. It is worthwhile pointing out several features. The most conspicuous feature is the difference in the summarized consumptive use. Since all plant species were grown under identical soil-, nutrient- and water supply and even meteorological conditions (the experiment was performed with all plant species at Szarvas in 1966), it was obvious that differences in water requirement were due to differing biological and physiological properties.

What have been told above are even more confirmed by the curves of Fig. 6 which represent the daily evapotranspiration of various plant species (Winter wheat, potatoes, sugar beet, mixed grasses) in the growing season of 1966. It is clearly seen that besides the meteorological factors the species (and on the basis of other investigations we can state that the variety too) and the plant phases also play a decisive role in determining the trend of water requirements and water use within the growing season. The maximum value of daily water requirement is different in each plant culture and its time coincides with the critical period of growth.

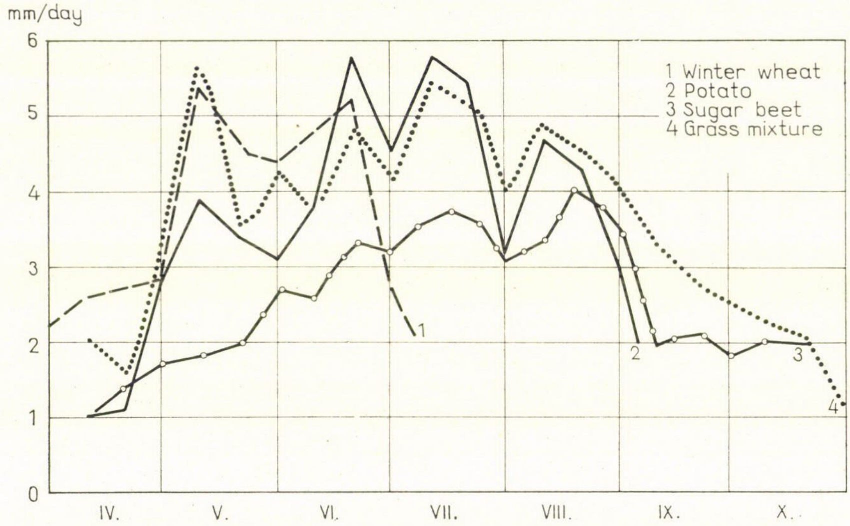

Fig. 6. Changes in the daily water use of different crops during the growing season in 1966 (Szarvas, medow clay, varieties: winter-wheat Bezostaya; potatoes Kisvárdai rózsa; sugarbeet beta-poly 2; grass mixture)

According to the mentioned authors (Borka—Borka 1972) the water use was the highest in the second phase of the growth of the peach trees, at the time of stone hardening in all three varieties. According to our investigations the highest water use of field crops usually coincides with the time of flowering.

On the basis of Fig. 6 it can be established further, that the character of the dynamics of water requirement in the individual plant species is determined primarily by their growth rate, while daily evapotranspiration decisively by the meteorological factors. Breaks seen on the curves of the figure are caused by the rapid daily changes of weather rather than by changes occurring in the growth of plants, as reliably proved by the data of Table 1.

The establishment of the paper (Borka—Borka 1972) that in the diurnal course of the transpiration of peach trees some decrease can be observed around noon, since the intensive radiation, high air temperature and low relative humidity in those hours reduces the turgor pressure in the plant and upsets the water balance in it, cannot be supported by facts, only made likely by considerations. The statement can be accepted as true in cases when the potential evapotranspiration (PE) or "evaporating capacity" of the atmosphere surrounding the crops is higher than the water uptake- and water transport capacity of the plants. If the two cannot keep balance (e.g. very high PE or very low soil moisture), under the influence of a "vapour hunger" in the air all plants containing water are "compelled" to lose water until

Table 2

Diurnal change of net radiation (R) and latent heat flux (LE) of lavender (gcal/cm² hour), between 3rd and 27th June 1959, Tihany-peninsula

Hour	*a)* 22 days average		*b)* 7 clear days average		*c)* 7 cloudy days average	
	R	LE	R	LE	R	LE
00—01	—3.4	—0.5	—4.1	—0.4	—2.2	—0.5
01—02	—3.3	—1.3	—4.1	—1.3	—2.4	—1.0
02—03	—3.4	—0.6	—4.1	—0.6	—2.4	—0.4
03—04	—3.5	—1.2	—4.2	—0.9	—2.3	—0.2
04—05	—1.9	—1.7	—2.4	—0.7	—1.0	—0.2
05—06	4.7	—5.5	6.2	—6.9	3.6	—2.4
06—07	13.0	—7.8	17.4	—9.4	9.4	—6.0
07—08	24.1	—13.0	28.2	—15.4	17.4	—11.0
08—09	35.3	—21.0	37.6	—19.2	31.4	—19.7
09—10	42.3	—24.3	46.3	—22.5	32.2	—21.4
10—11	45.8	—26,7	52,3	—29,1	33.6	—22.4
11—12	47.3	—27.9	54.2	K 27.5	35.3	—23.8
12—13	49.4	—30.0	54.7	—26.7	37.3	—27.3
13—14	39.2	—26.0	49.4	—24.0	31.1	—26.4
14—15	34,7	—26.7	44.1	—29.9	22.7	—24.5
15—16	28.1	—26.1	32.5	—27.5	18.5	—19.4
16—17	21.0	—24.3	23.3	—27.6	25.0	—17.8
17—18	10.3	—15.8	10.5	—16.9	5.8	—9.3
18—19	2.0	—8.8	1.7	—10.2	1·5	—5.4
19—20	—3.0	—4.1	—4.3	—4.0	—1.6	—5.1
20—21	—3.4	—3.3	—4.4	—3.7	—2.5	—5.0
21—22	—3.4	—2.4	—4.0	—3.6	—2.5	—2.8
22—23	—3.3	—1.1	—4.0	—2.2	—2.4	—1.2
23—24	—3.3	—1.1	—4.0	—2.2	—2.3	—1.2
daily gain	397.2	0.0	458.4	0.0	294.8	0.0
daily loss	—31.9	—301.6	—39.6	—312.4	—21.6	254.4

the water retaining ability of the material in question is brought into balance with the "evaporating capacity of air". Since "evaporating capacity" (potential evapotranspiration) reaches its maximum around noon, it is rightly assumed that on hot, clear, dry, windy days about noon the "water transporting ability" cannot keep up with the "evaporating capacity" even in aquatic plants, with the consequence of a natural decrease in the turgor state.

In earlier investigations (ANTAL 1961) we also proved a similar phenomenon on sunny days in the diurnal change of evapotranspiration of lavender, although this plant is generally known as belonging to the drought tolerant species. On the Tihany-peninsula (Balaton-lake) the water use of lavender was determined from hour to hour by the energy balance method (ANTAL 1966). The results of a three weeks measuring in a flowering lavender are shown in Table 2.

In the table the positive sign means energy intake, and the negative sign energy loss for the crops (evapotranspiration can be obtained in mm water by dividing the values of hours by 59.4). On cloudless days between one and two o'clock p.m. the rate of evapotranspiration — as contrasted with the average diurnal change of 22 days — comes to a sudden stop, then decreases, until increasing again after 2 p.m. Accordingly, a double wave can be found in the diurnal change of evapotranspiration by the lavender plant, with a main peak at 2 p.m. and a secondary peak at 10 a.m., as proved by the authors of the paper (BORKA—BORKA 1972) too, with a measuring method different from that described here.

On the basis of our own measurements the establishments of the paper (BORKA—BORKA 1972) concerning this question are completed by the following. It is known that a considerable part of the evapotranspiration of surfaces covered with vegetation is provided by the transpiration of plants. In some authors' opinion — including Henrici in: SHANBHAG (1957) — at about 10 a.m. there is no further increase in the transpiration, since as a result of the higher intensity of light the stomata of plants become narrow, through which stomatic transpiration decreases. This is especially valid in the case of plants for which no abundant water supply is ensured. The lavender plants on Tihany-peninsula are exposed to a very intensive radiation in the noon hours (southern slope of 10—20°). Sometimes the hourly maximum radiation to which the crops are exposed even exceeds 80 gcal/cm^2. In the most critical period of the growing season, at the time of flowering (Table 2) the 20—40 cm deep soil layer cannot provide the necessary water supply. Thus, the assumption that the decline of the water use curve in Fig. 7 is caused by a reduction on sunny days of plant controlled stomatic transpiration seems correct. The results obtained by Gy. Borka and K. Borka have to be completed in this context by adding that this reduction of water use can only be observed on sunny days when the plants really have to protect themselves from increased transpiration in order to maintain a favourable state of turgor. On cloudy days, or on the average of a longer period (see Fig. 7), water use has an almost perfectly regular daily course.

As to the methods of measuring applied in the study (BORKA—BORKA 1972), we should like to add some notions to the results obtained with the thermistor point thermometer. One of the objectives of the studies performed in the peach crops was to compare the temperature of the leaves suffering from water saturation deficit with that of the leaves in the state of full turgor. According to the authors' investigations if the leaves contained sufficient quantities of "mobile" water for intensive transpiration, then the temperature difference found between the leaves exposed to sunshine and those in shade was a maximum of 1—2 degrees, while when the leaves suffered from a water saturation deficit (lack of transpiration), the difference sometimes even reached 5 degrees.

As it is known, the temperature of the leaf is the result of its system of heat- and water balance. If the exposure of two leaves to radiation and other meteorological factors is identical, it is obvious that in case of identical water losses their temperature will be almost the same (leaf temperature being a function of water balance). When one of the two leaves suffers from

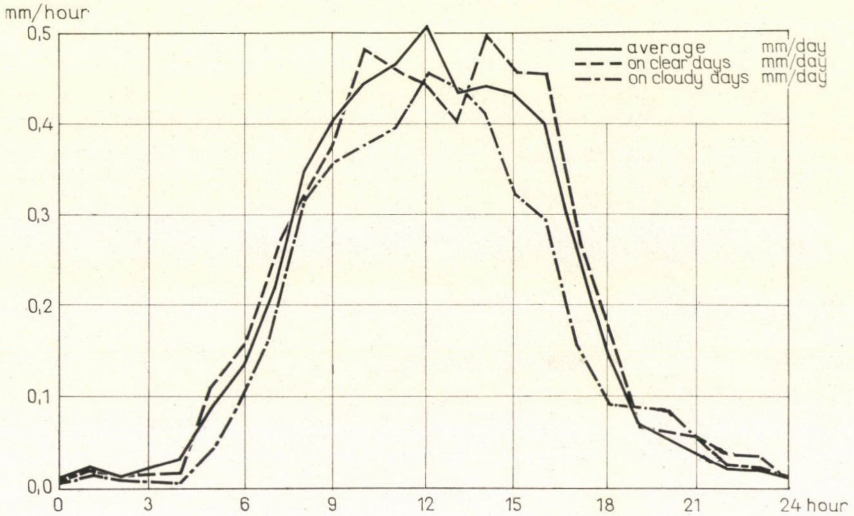

Fig. 7. Diurnal change in evapotranspiration of lavender (Tihany-peninsula, 3—7 June 1959)

water deficiency while the other is in a state of full turgor, then the temperature in the saturated leaf is lower. The leaf temperature difference is proportional to the difference in transpiration intensity, because the transpiration of each g of water involves the consumption of 600 gcal heat energy. And heat consumpting results in reduced leaf temperature (the leaf temperature changes induced by photosynthesis can in this case be neglected).

On the basis of the above mentioned the difference in temperature between water deficiency and water saturated leaves is easy to interprete energetically. To measure the leaf temperature difference (to be able to determine the influence of the water deficiency and water saturation of leaves have on leaf temperature) is, however, more difficult, as two leaves with perfectly identical exposure but considerably differing in water saturation are not likely to be found. If — on the other hand — the two leaves are differently exposed to the meteorological factors, then the differences in their net radiation may cause much greater differences in the leaf temperature than the differences in their water contents. Precise measuring can only be made when by adequate fixation (e.g. with forceps) a perfectly identical exposure (identical exposure to the sun, identical slope, identical height above the soils surface) is ensured for the two leaves, and the water supply is cut off completely for one of them. The leaf temperature differences measured in this case can be fully attributed to the difference in the water saturation deficit of the leaves until, finally, as a result of drying up, the albedo and with it the radiation absorption capacity of the leaf change.

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WHAT ARE THE INNER FACTORS INFLUENCING TRANSPIRATION?

The article by Gy. Borka and K. Borka sent to me for recension is undoubtedly interesting, since in it the changes in the water balance of three peach varieties are described in connection with the weather conditions and plant age.

Special attention has been paid to the changes in transpiration intensity; the changes in the general water supply, leaf temperatures and stoma behaviour are studied in relation to them.

It seems to me, however, that the article would have been more interesting, if the authors had paid more attention to the inner factors influencing transpiration, and first of all to the molecular dynamics of water. In this respect the interrelationship of only two water categories was investigated: the water extracted ("free") and that remaining in the leaves ("fixed") after the action of the water-extracting force. From the up-to-date viewpoint this interrelationship is only characteristic of the water holding capacity of the plant, which is dependent on a lot of factors. Among them the moving of tissue water has a great importance regarding transpiration.

The recent physical methods (dielectrical and infrared spectroscopy, nucleo-magnetic resonance, etc.) make it possible to measure the parameters (coefficients of self-diffusion, the time of molecule-relaxation), the thermal movement of water molecules are characterized by. By using these parameters which give the well-known conception of the structural make-up of water, first of all it is necessary to compare the course of the vital processes in plants.

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WHAT INSIGHT CAN BE GAINED INTO THE BEHAVIOUR OF PLANTS BY DISCUSSING "FIXED" AND "FREE" WATER?

The paper reports some observations on the water relations of peach trees. The main results fit comfortably within the current paradigm of water relations, although the discussion is in places confusing, or, at best, unhelpful. Thus, little insight into the behaviour of plants is gained by discussing "fixed" and "free" water; the generally smooth relation between the relative water content and the water potential of plant tissues means that the classification into "fixed" and "free" is completely arbitrary — it is much better simply to refer to water potential when comparing the water status of tissues. Again: it is misleading to claim that a leaf in the first storey "evaporated 46 per cent of the free water evaporation under the same climatic conditions, while leaves taken from the tenth storey evaporated 81 per cent"; this statement gives the impression that the canopy as a whole loses water several times faster than does a free water surface, which is absurd. The reader would be much less confused if the authors quoted stomatal resistances instead of fictitious evaporations.

The paper would be much improved if it were re-written after the authors had made a thorough study of a recent book on plant water relations such as that of Slatyer or of Kramer.

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CHRONICA

Dr Hollós László

LÁSZLÓ HOLLÓS*

1859—1940

Szekszárd, this charming little town is not only famous because of its great poet Mihály Babits but also a scientist known in Hungary only by those belonging to a narrow professional circle. This scientist, László Hollós was born in Szekszárd on the 18th June 1859. His family immigrated from Styria during the 18th century. His father who later became a defender of freedom as a lieutenant was born in Szekszárd in 1827, thus his family was rooted in this land. The environment of Szekszárd, the rich flora and fauna of the country, its picturesque scene inspired the later outstanding botanist and mycologist, as he himself wrote — not without lyric enthusiasm — in the preface of his extensive study published by the Hungarian Academy of Sciences:

"I did not even go to school yet when I used to slip away to the Szekszárd woods where I collected flowers and beetles. I ran after butterflies and sought for bird nests. Even my mother anxious for her only child, could not break my habit of frequenting the woods. Whenever there was a chance I ran off to the wood, and this love of the wood remained with me during my whole life. As a man I often turned my back to the dusty town, went to the woods and took delight in nature."

Thus wrote the 74 years-old mycologist renowned the world over in his last — hundredth — work, who, though spending his life far from the public in the clear atmosphere of

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* Lecture delivered at a commemoration held in Szekszárd on October 12th, 1970.

science, was not free from troubles, which — however small they seem for posterity — made his life tragic. His life was also tragic because his great dream to come to Budapest, to the centre of mycological research was unattainable; the post at the Botanical Department of the Hungarian National Museum was given to Gusztáv Moesz, a younger researcher who later worked with affection on clearing up the taxonomy of fungal species in the vicinity of Szekszárd described by Hollós, and after the latter's death wrote a very human and objective necrology in the Botanical Bulletin of Hollós, the scientist and man. If not friendship but a mutual appreciation developed between the two great Hungarian mycologists of the 20th century, who — we can say — did the most in exploring the Hungarian flora of fungi. Their names appear most frequently in the international literature too. In the mentioned necrology G. Moesz wrote the following about Hollós:

"Mycologists who did not know him in person, when hearing his name will think of his masterpiece 'Magyarország Gasteromycetái' (Gasteromycetes of Hungary). His name became known all over the world by this work. Those who knew him in person saw not only the prominent scientist but the human being too. Some phases of his life showed him as a scientist, others primarily as a human being. His prominent works: 'Magyarország Gasteromycetái' (Gasteromycetes of Hungary), 'Magyarország földalatti gombái' (Underground fungi of Hungary), 'Kecskemét vidékének gombái' (Fungi in the district of the town Kecskemét), 'Új gombák Szekszárd vidékéről' (New fungi from the vicinity of Szekszárd), 'Szekszárd vidékének gombái" (Fungi in the district of Szekszárd) were undoubtedly the results of the untiring diligence, enthusiastic love of work and knowledge of a scientist. We wonder, however, whether the destruction of his much loved fungi, an important collection and work of many years in 1911 was also the act of the scientist? This rash destructive work was certainly not done by the idealism of the scientist, but by some slip of the human soul which could be explained correctly only on a psychological basis . . ." The writer of the necrology here alludes to the fact that in Kecskemét where he had taught for 20 years and rendered unforgettable service by writing several famous papers, tragic events took place in 1911. It happened that two talented persons with highly different ambitions met and clashed for one second: Pongrác Kacsóh, composer of the opera János Vitéz, already a celebrated artist, who was appointed provisionally to succeed the deceased István Hanusz as director of the secondary school wanted to use Hollós' botanical department as a concert hall to instruct the youth in music. Hollós did not understand the director's intentions and — though Kacsóh soon left the post which was not his real field — could no longer remain. On 16th July 1911 he retired and lived at Szekszárd, where he had been born, till the end of his life. All this was tragic in itself, as he gave up scientific work for a long time, did not teach and, no doubt, deprived the Hungarian literature of many important mycological works. But the greatest blunder of Hollós was that he burnt and buried his renowned mycological herbarium and other collections which in his life he had bequeathed to the Botanical Department of the National Museum. Thus it happened that the most famous Hungarian collection of fungi, the rich material collected by a diligent scientist was destroyed, which is a great loss for certain groups of the taxonomy of fungi, such as Gasteromycetes and Fungi Hypogaei.

Hollós' career was not unbroken, but his scientific work is a rock of granite which time and weather cannot destroy; it erected a worthy monument for a modest, enthusiastic creative man who was not always devoid of human faults. He wrote about a hundred compositions and papers, not to mention his attempts with fiction published under the assumed name Corvinus, which he himself did not consider important. 59 of his works are on mycology including the excellent monographs dealing with the whole system of fungi, such as: "Kecskemét vidékének gombái" (Fungi in the district of Kecskemét) and "Szekszárd vidékének gombái" (Fungi in the district of Szekszárd) published in 1913 and 1933 respectively, under the auspices of the Hungarian Academy of Sciences. He wrote 9 works on floriferous plants, 1 on ferns, 6 on geology, 2 on chemistry and 4 on archeology, proving his wide scope of interest and thorough knowledge. Beside his most prominent work "Gasteromycetes of Hungary" (1903) published also in German in Leipzig (Die Gasteromyceten Ungarns, 1904), it is the two above mentioned monographies of summarizing nature that make Hollós' activity actual and exemplary. These works provide evidence of his amazing diligence too. In his large floristic work he pointed out and specified 1934 fungus species from the Kecskemét district from the Szekszárd district he collected and specified 1386 fungus species with a number of new species included, though the latter had to be revised by G. Moesz. He wrote 12 publications on fungi observed in Kecskemét and Szekszárd. The number of new species introduced in science by him is about 480 which points to a very remarkable and successful activity. It is interesting that most of the new species belong to microscopic fungi, mainly to the taxonimic groups of Ascomycetes and Deuteromycetes (Fungi imperfecti). Of this work he himself writes: "I did not seek for these new species purposefully but found them while searching for the truth. And if some of them will later prove to be already known — (as it happened, see Moesz, Botanikai Közlemények, 1928, 1929) — I had the pleasure — while investigating them — of forgetting the everyday struggles in the clear atmosphere of science."

His life was concentrated round two foci. One of them was Kecskemét, where during his 20 years activity as a teacher he not only taught generations of students to love nature and honour good and noble things, but also laid down the foundation of his work on mycology. His greatest and most lasting works were also written there, at that time. The other focus was Szekszárd, his birthplace, where he learned to decipher nature's book and then to read it fluently; and where after the sad episode he regenerated his passion for life and took his pen and microscope into his hands, and in his advanced years, perhaps as his swan song wrote his large mycological monograph of Szekszárd, perhaps more flexibly, with practical life taken more into consideration; as if Szekszárd had drawn the man and scientist from the too idealistic atmosphere of science back to everyday life. Many of his works (e.g. Ramularia, Septoria, etc.), minor monographs and large floristic works (Kecskemét, Szekszárd) contain phytopathologically important Hungarian species. Thus Hollós' activity was essential for the Hungarian phytopathological research too.

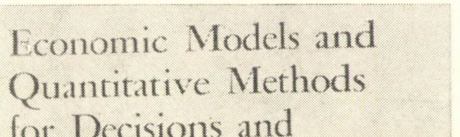
A short commemoration cannot give a detailed appreciation of the lifework of a great scientist. Therefore permit me here to bow my head in admiration before László Hollós, the renowned mycologist, on the thirtieth anniversary of his death. He fought his way to world fame and drew the attention of scientific circles in Hungary too to himself. In 1904 he became a corresponding member of the Hungarian Academy of Sciences, thus he got to the summit where scientific merits are acknowledged above all. Hollós never sought for success, he represented the modest quiet type of scientist, and it seems more worthy of his memory if we casually mention his scientific merits and see in him one of the greatest Hungarian mycologists and set him as an example to be followed by the young generation of scientist. We can serve the memory and life work of László Hollós best if at last we call into existence the modern Hungarian centre of mycological research, and continue the exploration of mycology in Hungary where Hollós deceased 30 years ago in Szekszárd.

G. Ubrizsy

Acta Agronomica Academiae Scientiarum Hungaricae, Tomus 21 (3—4), pp. 475—484 (1972)

RECENSIONES

Economic models and quantitative methods for decisions and planning in agriculture. The material of the International Seminary held at Keszthely, Hungary, in 1968. Proceedings of an East-West Seminar. Editor: Earl O. Heady. The Iowa State University Press. Ames, Iowa, 1971, 518, XIV.



Economic Models and
Quantitative Methods
for Decisions and
Planning in Agriculture

Czechoslovakia
France Germany
Hungary Italy
Netherlands Norway
Poland Romania
Sweden Switzerland
USSR United Kingdom
United States of
America Yugoslavia

Proceedings
of an East-West Seminar
edited by
EARL O. HEADY

The bulky volume gives a full review of the session held by the internationally best known agricultural economists and business economists on the subject of applied economic mathematics at Lake Balaton three years ago. The uniform conception reflects first of all the thoughtful and competent editorial work of Curtiss Distinguished Prof. Heady, of the Iowa State University, U.S.A., Honorary Member of the Hungarian Academy of Sciences. The relatively long time the American edition took to get through the press hardly did harm in this case to the actuality of the content, moreover, the mature and better integrated formation of not only the comments but also of the reports and the evaluations introducing a discussion make it easier for the reader to get the information required by him.

In the mid fifties the agricultural economists of many countries in the world showed an increasing interest in applying economic-mathematical models and methods in making decisions and preparing plans in agriculture. Following the theoretical researches of a pioneer character, practical attempts were made in an increasing number at all three levels of the economy, in the micro-, mezo- and macroeconomy. The efficient econometric and operation research methods were employed in the new approach to the problems of farms, groups of farms, entire districts and national economies. It can be stated without exaggeration that methodological schools developed around certain great personalities of this field of science, mainly under the influence of their teaching and literary activities, which went beyond the borders of their countries from the beginning. Progress was not and could not be without contradictions; the conflictng views met more and more frequently with fruitful discussions and debates. It was such preliminaries that contributed to the appearance of a demand for convening a seminar to assess the situation, sum up and evaluate the results obtained and explore the possibilities of development.

Under the chairmanship of Prof. Heady an International Preparatory Committee was organized with Michele de Benedictis Italian, Vladimir Kadlec Czechoslovakian, Kravtchenko Rosztislav G. Soviet, Danilo Pejin Yugoslavian, Ulf Renborg Swedish and József Sebestyén Hungarian professors and research officers, respectively, as members. During its one-year functioning the Committee did a thorough work preparing a well-considered integrate work program, as reflected by the table of contents of the book. The chapter headings are: I. Foundation and background in planning models. II. Problems and potentials at the micro level. III. Regional models of planning and development. IV. Experiments and experiences with national planning models for agriculture. V. Formulation of national models. VI. Gaps between plans and realization and practical possibilities for improvement in performance.

The chapters listed above are the summarized subjects of the consultations at the Seminar, and after careful considerations the Committee invited the most competent experts of the given subjects to present a summary of them or make the first critical comments to begin with the debate. The efforts were, in general, successful, as proved by the content of the book. The manuscript of the lectures was available for the participants of the Seminar in advance. This preparatory work ensured the adequate level of the discussions.

It is to the Hungarian Academy of Sciences — as acknowledged several times in the past years and also in the preface of the book — that we owe thanks for inviting the Seminar to the Hungarian People's Republic, and with its high reputation and apparatus giving assistance in creating the conditions of an efficient cooperation at the consultations. An important part was played in the work by the Keszthely College (now University) of Agricultural Sciences and the Research Institute for Agricultural Economics, Budapest. The fact that the book contains reports from as many as nine Hungarian participants — which in proportion competes with the role of the most developed countries — proves the relatively advanced stage of Hungarian research work and the high performance of experts apart from the advantage following from the position of the host. There was one Hungarian report to every five foreign reports.

It is hard to read the words of welcome of the late Ferenc Erdei, then vice-president of the Academy, without being moved. He dealt with the perspectives of improved management and methods in agricultural economics. It was in relation to the great perspectives — the revolutionary progress of science, its becoming a force of production, the conquest of nature, the fight against hunger and in order to provide sufficient food for the whole human nation that he appreciated the importance of consultations and of an international cooperation realized in them. He pointed to the importance of a wide practical application of well-proved efficient methods. The book published recently is at the same time a document of his judgement having remained valid.

Heady's introduction and first report dealing with the problems of a synthesis between the economic environment and the means of decision making and planning are outstanding. The basic question is whether in different socio-economic systems the same methodological procedures can be applied to express economic considerations in planning, and preparing and making decisions. It is on the answer given to this question that it depends whether there is any use and reason for the existence of an international scientific cooperation in elaborating, implementing and improving the economic-mathematical methods and models. The report specifies the elements on the basis of which it gives a positive answer. Planning — at least on an operational level — plays an important role in every country of the world. Farms operate everywhere under conditions of scarcely available resources and other restrictive factors. While different economical systems define the tasks of agriculture and agricultural enterprise differently, some object is set everywhere, and this object can be expressed in a simplified form by an objective function. Competing or replaceable branches and technologies are similarly usual in agriculture. A considerable part of the elements can be defined numerically; planning and decision making even call for quantification. Heady has realistic views, does not hide the differences, and in the knowledge of these differences and acknowledging them he tries to create a synthesis — within the limits of generalization — not only in connection with the socio-economic systems but as regard to the economic spheres too. He follows up the interactions manifesting themselves in the methodological development of enterprisal, regional and national economic models, emphasizing the analogous elements. This clue is followed up in the comments made by the Polish professor Rychlik to introduce a discussion.

The different nature of socio-economic systems is reflected in the composition of the book too, apart from their theoretical treatment. In each scope of subject at the Seminar, work was divided in such a way, that either the general report or the comments of the first discussant were made by a representative of a socialist country, while the other task was performed by a scientist of one of the capitalist countries. Thus the authors listed in the table of contents are distributed among the two world systems — or with the term used by the title of the book: between West and East — in almost equal proportions. Of the fifteen countries represented at the Seminar six were socialist countries, and from the 63 foreign participants 38 contributions have been included in the publication. This readiness to debate clearly shows the many-sided international interest in the subjects.

In the first chapter reports from Baker and Tintner American, and from Edemsky and Aidin Soviet experts, as well as comments made on them by Polish, Hungarian and Yugoslavian participants survey the methods and procedures, and make attempts to systemize them, and by presenting their occasional application, e.g. under the conditions of economic development in the developing countries — find out what kind of approach is the most suitable to solve problems of different character (linear and non-linear, static and dynamic, welfare and growth, etc.). The theoretical arsenal of which this chapter gives account — from the traditional planning based on calculations to the most sophisticated optimizing and simulation models and models systems — is linked with many threads to the most recent results of computer techniques. The material of the Soviet research workers is especially interesting, as they employ a mathematical method in determining the possible variations of linear economic-mathematical models and selecting them according to the given problem, as a function of index number-groups, conditions-groups and optimum criteria. The whole chapter is interspersed with the authors' naturally differing preferences of the various solutions; expertness influences the judgement and this gives an opportunity to expound the opposing arguments.

The authors of the summarizing reports in the second chapter dealing with the operative planning and decision making are: the Swedish Renborg, the West-German Reisch and Weinschenk and the Soviet Kravtchenko professors, while the discussion is introduced by papers from Hungarian, Yugoslavian, English and Roumanian participants. The so far abstract treatment of methodology is completed here by an analysis of questions and difficulties more closely connected with practical application. Beside a number of important partial problems the reader will often find analyses of correlations of economic environment and farm, of assumptions ensuing from it, of ensuring the initial farm data and organizing their collection, of models made for farm groups and of the development of adequate farm groups, etc. During a discussion about the organization and levels of decision and information the system aspect and the cybernetic schematization are given a role too. The different methodological requirements of long-, medium- and short term planning as well as of operative planning, and as to the latter the application of network diagrams are referred to by many. The advisory service in the agriculture of the capitalist countries where family farming is in preponderance, and in the socialist countries the internal and external controlling organs of state- and cooperative farms and integrated agricultural enterprises wish to rely to a greater extent on the possibilities offered by the new methods, especially on the quick massive calculations of computers. Planning and decision preparation in integrated and large size farming units, agricultural enterprises require a complex model system, the structure and inner correlations of which make a many-sided theoretical approach necessary. The discussions make it clear that the possibilities of applying the mentioned up-to-date methods are more favourable under socialist conditions, and in a market mechanism controlled by more permanent economic regulators their usefulness may also increase.

The development of regional planning and decision making models is somewhat different in direction and dimensions from that of the farms. Large regions of developed economies or regional undertakings of international interest encourage the application of up-to-date quantitative methods and the construction of models. The third chapter of the book contains two American (Swanson, Hall-Heady), two Soviet (Mash-Kiselev, Miloserdov) reports, and a French report from the apparatus of the International Bank for Reconstruction and Development (Oury), completed by Hungarian, American, Swiss and West-German comments introducing the discussions. The regional models are equally connected with the preceding and following subjects of the Seminar, because they can partly be built on a large number of representative farm models, and partly — perhaps through interregional traffic or production allocation problems — serve as precedents for national models, lead to them and make their more differentiated development and allocation possible. Their object- and condition systems develop according to their peculiar features, their relations to certain branches within the economic environment — e.g. to food industry, for reasons of supply — are given a greater emphasis. (This is a remarkable approach to the concept of food economy and to the opinion that on the mezo-level the problem of branch models cannot be neglected either.) From the point of view of mathematical methodology, non-linear and regression models are discussed more often in this chapter.

The fourth and fifth chapters of the book can be reviewed together, since they both deal with the national level, macroeconomic sphere. The reports of the fourth chapter give account of national models at different stages of preparation from Czechoslovakia, France, Yugoslavia, Hungary, German Federal Republic and the United States of America. The greater part of the models described start from a highly simplified form and try to refine it in successive steps. The specific agricultural models are generally adjusted to national models covering the whole national economy. Owing to the high number of variables and limiting factors included, more complex models can be handled only with matrices of an ever increasing size. Discussion about the macro-models was started by a single Yugoslavian contribution. The subject of national planning in agriculture is introduced by the joint report of Kálmán Kazareczki and József Sebestyén (Hungary). Further reports were held by Polish (Herer-Porwit) and French (Tirel) participants, with English and American comments to introduce the discussions. The main subjects of discussion were: priority of planning objectives, interrelations of branches, forecasting of expected future trends of certain factors as required for quantification. The reports touched upon the development requirements and -trends of the methodology of dynamic solutions. Emphasis was laid upon the adequate relation of agricultural policy and agricultural economics in national planning, and upon the importance of a decision maker being able to choose between alternatives.

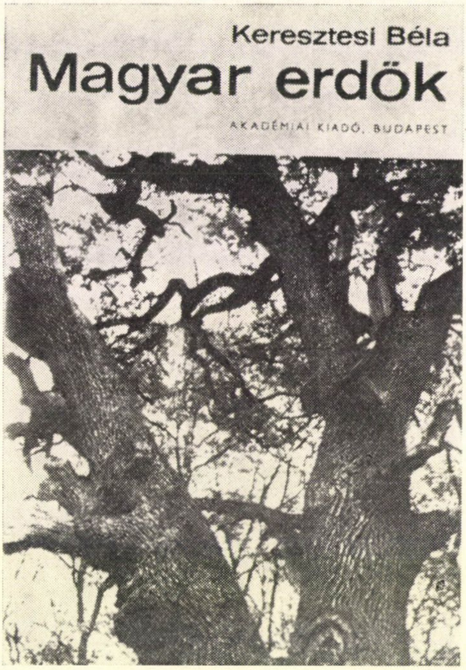
The authors of the reports published in the last chapter of the causes, extent and solutions of differences between the plans and their realization where Hungarian, Norwegian, Czechoslovakian and Dutch (Kovács, Reisegg, Eremias and de Veer) participants, and Italian, American, Hungarian and French experts took part parallel as first discussants. This chapter presents a wide range of experiences, observations and recommendations concerning the application of the methods discussed during the Seminar, with the aim of promoting their wide and efficient distribution. The reports deal with the question of an adequate training of experts and farm managers as a precondition. Theoretical development and practical interest should be coordinated. The methodology can be refined by a simultaneous and continuous improvement of the activities performed in the three economic spheres.

Unfortunately, the material of the detailed discussions could not be included into the book, not even as a summarization according to subjects. On the other hand, the accurate index annexed to the book offers a great help to the reader in using the volume. The references are in connection with the text of the reports. The almost 450 works listed constitute the summary of the essential bibliography of this field of science.

The few pages information on this vast material is — of course — able to give but the outlines. Unfortunately, there are only a restricted number of Hungarian readers who possess sufficient knowledge in English special language to read this book, or study the parts they are closely interested in. We have been informed about an edition in the Russian language expected in the near future in the Soviet Union, in keeping with the agreement reached at the Keszthely Seminar. It would be useful if a Hungarian translation of this collection which gives a comprehensive view of this filed of science were available.

Gy. Radovics

B. Keresztesi: *Magyar Erdők* (Hungarian woods). Akadémiai Kiadó, 1971, 432 pp. 470 figures.



In the preface of the second edition of his book the author mentions that the first edition brought an unexpected success, thus, within a very short time, the Publishing House of the Hungarian Academy of Sciences published an enlarged second edition.

I had the honour to review the first edition of Keresztesi's book in the Acta Botanica. I wrote with appreciation that the literature on forestry had been enriched by a remarkable new colour. In addition to the author's modern conception, the book with its wonderful style and pictures selected with apparent aesthetic sence offers a delightful experience not only to the experts of forestry but also to those interested in natural sciences. Beside the economic importance of forests the book points to values of no negligible significance.

The second edition is still more praiseworthy. The author follows the changes occurring in the human mind when recognizing and appreciating the values of natural environment. The remarkable fundamental idea of the book is related to the world-important long-range intergovernmental and interscientific research conception of Man and biosphere. The author considers the forest with its various colourful and useful aspects as an important environmental factor of human life.

It is refreshing to read how the professional forester besides the profit also notices the aesthetic quality of the forest and its influence on the healthy functioning and general condition of man.

The new part of the enlarged edition deals with the introduction and evaluation of wild life, and shooting in Hungary.

In the first chapter the author discusses the "Manysided utilization of the forest". He emphasizes that besides the industrial utilization forests are exploited in many respects all over the world withvery few exceptions. He supports this statement with a number of foreign examples.

It was Endres who introduced the concept of "The public welfare effects of the forest" at the end of the last century. By this term he meant the effect of the forest on climate, water management, agriculture and the prevention of natural disasters, as well as its sanitary and ethical aspects. It is not useless to mention this interpretation when presenting the book, as this scope of ideas very strongly impresses the work.

The outstanding merit of the book is that it presents the many-sided correlations of man and forest scientifically but at the same time vividly, thus calling attention to the various characters of the forest. The chapter brings the man of this busy world who loves nature into close contact with the forest when it points to the relation between tourism and natural science.

Under the title "The aesthetics of the forest", through the examples of Hungarian forests developed in regions of different geographical character the author shows what a harmonious aesthetic experience a forest stand may offer by the various colours of flowers, fruits and foliage. This harmonious unity includes all being — the whole multicoloured and multitone biocoenosis.

Where the author thinks that the prosaic words do not adequately express his feelings, he provides for the emotional elements by interjecting passages of poetry. Besides presenting the beauty of forests, he refers to the individual aesthetic effect of some tree species (white poplar, Scotch fir, black fir, spruce, beech, etc.).

The work presents "Forest scenery" as a category of natural geography. The author emphasizes that the ancient forest scenery is very rare, even when considering the whole world, we must therefore appreciate the slightly modified natural landscapes where the intervention of man has disturbed the harmonius biocoenosis of the forest to a minimum extent. The concept of forest scenery is treated from the points of view of sanitary, cultural and aesthetic education.

Within the subject of "Hungarian forests", after a survey on the natural geography of forests economic questions are dealt with and a critical evaluation presented. The important sporting- and economic aspects of wild life are dealt with in a separate chapter which increases the number of those interested in the subject. In the course of a detailed presentation of Hungarian forests, those of the Great Hungarian Plain, the lower part of the Alps, the Transdanubian Hills, the Transdanubian Mountains and the Northern Medium-Height Mountains are discussed separately. Besides its emotional effect the chapter contains useful information for those who love nature.

The chapter entitled "Welfare forest management" discusses the aspects of preserving and increasing the influence of forests on public welfare apart from their economic utilization. The author presents the aspects of planning and the aesthetic features of welfare forest management. He deals with the landscape aesthetics of afforestation, and touches upon the question of protecting old trees, tree-groups and alleys which have scientific value.

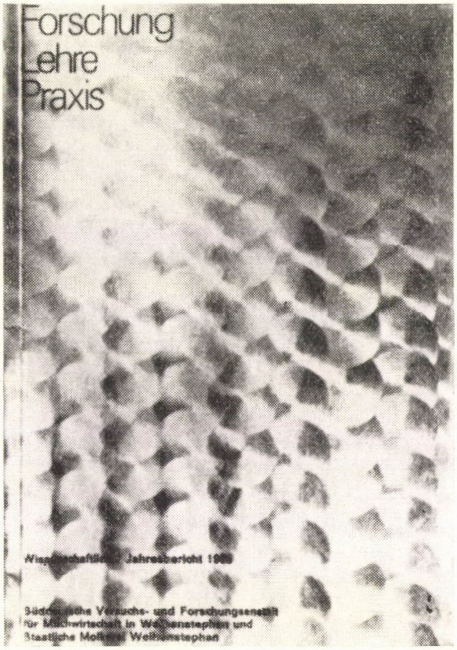
The author mentions that the establishments in forests (tapped springs, fire laying places, shelter huts, ranger lodges, etc.) should be shaped in harmony with the landscape and offer a picturesque view for those who settle down at such places.

In the last part of the book the author deals with the investment policy of forestry which observes the aspects of welfare forest management.

Keresztesi's book with its 432 pages and 470 artistic photos is of interest not only to experts. Its social effect is manifested by its pointing to the multilateral connection between man and forest, its awakening the love of the forest and thus involuntarily promoting its protection.

The illustration is equal to the professional and aesthetic content of the book.

I. Kárpáti



Forschung, Lehre, Praxis, 1969, 1970, Süddeutsche Versuchs- und Forschungsanstalt für Milchwirtschaft in Weihenstephan und Staatliche Molkerei Weihenstephan

In August 1970 and July 1971 the South-German Institute for Milk Research and -Test led by Friedrich Kiermeier published in Weihenstephan reports on the 1969 and 1970 activities of its own, the Milk Section of the Munich Technical College and the State School of Milk Farming on 95 and 96 pages, respectively, under the title Forschung, Lehre, Praxis.

In the prefaces the editor refers briefly to the work done and difficulties encountered by the institutes in the previous years, underlining that their activities are of an educational, research- und practical nature.

Major activities of testing and research:

1) Aflatoxin formation in cheese

The investigation aimed at obtaining more thorough information on the toxic substance formed in pure cultures of *Aspergillus flavus* Link, *Aspergillus parasiticus* Speare and *Penicillum puberulum* Bainer on uniform culture media. Aflatoxin has four fractions of which fraction B_1 is the most important one. Fraction B_1 was examined by extracting it from the mycella, isolating it by thin-layer chromatography, measuring it with spectrophotometry; and all this was used as an "applied method" in cheese tests. 169 cheese samples — mainly Tilsit, Edam, Romadur and Camembert — were examined.

The results of the examination are:

a) Camembert cheese displays noble rot and does not produce aflatoxin, moreover, it prevents the fungi producing aflatoxin from growing.

b) In Romadur cheese fraction B_1 of aflatoxin occurs even in the case of inhibition by noble rot, but if a greasy texture has developed, this is generally excluded.

c) With a relative humidity of 79—99 percent the B_1 fraction of aflatoxin is produced on the surface of Tilsit cheese at temperatures of 18—30°C by *Aspergillus flavus* and *para-*

siticus, while at 5—18 °C by *Penicillum puberulum.* Under optimum conditions (temperature: 26 °C, relative humidity: 99 percent) when *Aspergillus flavus* has overgrown the surface of the cheese, the B_1 fraction of aflatoxin may be formed in 3—4 days. With the above fungi present, the formation of the B_1 fraction cannot be exluded in the cheese factory.

2) Copper content of milk and milk products.

The copper content of milk and milk products was demonstrated with 1,5-diphenyl-carbazone used as reagent. The biologically required copper content is 0.04 microgramme, while the average copper content is 0.56 mg/kg dry matter content of cheese, with a deviation of 0.2—3.4 mg, the first grade of Gauss' distribution frequency. Copper content was significantly higher in the Gruyère cheese than in other kinds of cheese. The average was 17.3 mg/kg, with a deviation of 3.8—27.0 mg/kg, the second grade of Gauss' distribution frequency. Chese prepared in a steel boiler contained less copper. The 0.6 mg/kg average copper content of Camembert cheese only inhibits the processes of lactous fermentation and volatile acid formation slightly.

Inhibition by copper is higher during the decomposition than during the synthesis of l-lactate, and higher during the synthesis of l-lactate than during that of d-lactate. In the Gruyère propionic fermentation was inhibited by the copper content to a greater extent than lactous fermentation.

Literary data as to the development of flavour and taste are contradictory.

According to the investigations of the institute diacetyl and acetoine originate from pyruvic acid, and their quantities increase in the presence of copper.

According to some data Gruyère was found to have a better taste when made in a copper than when prepared in a steel boiler.

In cheese with a metallic taste owing to a copper content of 20 mg/kg, differences in the products of protein decomposition as compared to the control could not — even with electroforesis and chromatography — be demonstrared.

Copper inhibits the activity of the original alkaline phosphatase-, β-glucoronidase-, peroxidase-, ribonuclease- and xanthynoxidase enzymes in milk.

3) Milking disturbances of animals giving milk.

When studying mastites, quarter udder samples were taken, coulter counter, Schalm-test and bacteriological examinations applied, and it was found that examinations on the basis of cell contents are the most suitable for diagnosing mastitis.

Staphylococcus aureus shows higher pathogenity than streptococci do. In the course of quarter udder examinations in 70 percent of the cases no differences were found in the 500,000/ml cell number of the first streams of milking (in the milk of healthy animals). In accordance with earlier studies it was found that the original enzyme activity of milk was higher in animals suffering from mastitis than in healthy animals.

A new result of the examinations was the relatively low correlation coefficient found between lactoperoxidase activity and leucocyte-count.

4) Synthetic materials in dairy industry.

When studying the physical and chemical reactions of polyethylenes they were found to aoutooxidize and produce peroxides, alcohols, carbon acids, ketons and their combinations. The main points of attack in the process are the double bonds of the carbon cycles. Oxidation also occurs under the influence of heat and UV radiation.

Ion exchanging processes also occur between the milk and the synthetic materials. As a result of the processes of oxidation hydrocarbons (e.g. ethane, propane, butane), carbon acids (e.g. myristic acid of C_{19-21}, C_{24-26}, C_{38-41} carbon number), methyl esters and other decomposition products can be demonstrated and it is they that cause the special "taste of synthetic material".

From the PVC tubes of the milking machines the monomer plasticizers are dissolved by the detergents, and fats enter the synthetic material, too. The B_2- and

C vitamin content of milk packed in synthetic material decreases under the influence of light with a simultaneous increase in the thiobarbituric acid-number.

5) Coagulating enzyme in sterile cocoa milk drink.

Cocoa powder was found to exert a coagulating effect of enzyme origin in cocoa with milk. The enzyme acts on the χ-casein, and appears together with the hear- and acid-induced coagulation. In ultra high temperature heated milken cocoa this enzyme does not become reactivated and thus does not cause coagulation.

6) Interaction between the original enzymes of milk and the insecticide substances.

During the examinations it was found that the organic phosphatase esters exert a non-competitive inhibition on the activity of xanthinoxidase, and a competitive inhibition on that of the alkaline phosphatase.

Constants determined by Michaelis for some insecticides:

p-nitro-phenyl-acetate:	7.6×10^{-3} mol/l
diaethyl-paraoxon:	1.9×10^{-4} mol/l
di-n-propyl-paraoxon:	2.1×10^{-4} mol/l

Order of the intensity of inhibition: di-n-propyl- > diaethyl- > dimethyl esters.

7) Changes in fatty acid esters in the presence of chlorinated hydrocarbons.

The presence of chlorinated hydrocarbons was found to exercise a considerable influence on certain phases of auto-oxidation in fatty acids.

8) Residues of quaternary ammonium compounds destroying the useful flora of milk.

Quaternary ammonium compounds widely used in the dairy industry when getting purposefully or accidentally into the milk considerably inhibit the fermentation activity of the useful flora of industry, thus unfavourably influencing the technological processes of cheese-, butter- and sour milk products. The literature has raised the problem of how these compounds could be demonstrated specifically, quantitatively and qualitatively in the producers' fresh milk. The Institute has developed a technique in the producers' fresh milk. The Institute has developed a technique in which an eosine-quaternary-ammonium bond is first created which can be demonstrated with 1.2-dichloroethane, by photometry at 542 nm wave length.

The limit, it can be demonstrated at, is 0.05 mg/l in aqueous solution, and 0.5 mg/l in milken medium.

9) Experiments with useful animals

In the course of the experiments the possibilities of increasing the yield were investigated with pigs, cows and fattening cattle. With animals producing milk the main object is to increase the yields of milk, protein and fat.

Climatic conditions as well as pasture- and feed management are dealt with too.

With insemination the deep freezing technique while in the case of male-tests progeny control is applied. The biological section of the Research Institute deals with the determination of factors controlling the formation of milk, with the synchronization of the cycle, studies the adrenal activity of the foetus; further, in the course of demonstrating gestagen substances (e.g. chlormadinonacetate) in milk it was found that 75 mg chlormadinonacetate applied parenterally could be traced in milk, while 10 mg could no longer be traced. In the faeces and urine of calves oestrogenic substances, such as, e.g. the diethylstilboestrol compounds, can be well demonstrated both chemically and biologically.

The method used at the Institute is suitable for the purpose of examining milk products, especially milk powder, which is important from the point of view of export.

10) Experiments on cheese preservation

Storage experiments were carried on for fourteen weeks at temperatures between 10 and 20 °C. Before storage the various kinds of cheese were dipped into a solution containing 25 percent potassium sorbeate.

3.1 percent of cheeses treated and 97.7 percent of those untreated were spoiled during the experiment. The cheese paste, when poured, contained 0.006 percent potassium-sorbeate.

50—240 mg/100 g sorbic acid and 40—

125 mg/100 g pimaricine, when present in cheese, totally inhibits mould formation for 40 days.

The food sanitary importance of the two mould inhibitors consists of their considerable inhibiting effect on the formation of afflatoxin in cheese. This toxin may penetrate into the cheese even to a depth of 1.6 cm.

11) Analytical studies

The components of milk: fat-, protein- and sugar content were examined with infrared IRMA II milk analyser at ± 0.033, ± 0.025 and ± 0.020% exactness. As a reference study, Gerber's fat-, Kjeldahl's protein- and Luff's sugar determination methods were applied with deviations pf ± 0.06, ± 0.08 and ± 0.07 percent.

12) Bacteriological examinations

The Institute deals with the question of postinfection, performs coli-titer determinations, Pseudomonas identifications, carries out studies on lactic acid, streptococci and enterobacterial metabolism.

Coli-titer, the determination of the total number of germs is made by the principle of the most probable number (MPN).

For the identification of *Pseudomonaceae* glutamate-starch-penicillin agar, oxidase-test, cultures kept at 4 and 41 °C, lecithin decomposition, gelatine liquification, trehalose-, mannit-, inosite-, hyppurate-, p-hydroxy-benzoate-, tryptophane-, phenyl-acetate-, benzylamine-, acetamide decomposition are used.

The metabolism studies of *Enterobacteriaceae* included the fermentation products of glucose.

In addition to the above, the Institute carried on milk purchasing, dealt with the problems of milk prices, performed market research and planning.

When studying the books, we find that from the data presented in them the work done by the institutes in 1969 and 1970 can be sufficiently evaluated. On the basis of two years' literary activity and dissertations submitted and accepted the data can be reproduced.

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